Spring 2012

The Effect of a Bcaa Supplement with and without Cho on Performance in Recreationally Trained Cyclists

Alaine Carleton Mills

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THE EFFECT OF A BCAA SUPPLEMENT WITH AND WITHOUT CHO ON PERFORMANCE IN RECREATIONALLY TRAINED CYCLISTS

by

ALAIN MILLS

(Under the Direction of Amy Jo Riggs)

ABSTRACT

The purpose of the current study was to examine the effect of a branched-chain amino acid (BCAA) supplement with and without carbohydrate (CHO) on performance in competitive cyclists. Subjects were recruited from the GSU cycling team (n=6). Each subject’s maximal oxygen uptake (VO2 max) was determined on an electrically braked cycle ergometer using a graded exercise test. In a randomized, double-blind, crossover design, subjects received one of three supplements prior to exercise: BCAA-1 teaspoon BCAA powder (Optimum Nutrition® Instantized BCAA 5000mg Powder containing 2.5g leucine, 1.25g isoleucine, and 1.25g valine), mixed with 500 ml of a zero calorie sports drink (Powerade Ion 4 ZERO®- Orange, containing potassium, sodium, and vitamins B12, B3, and B6); Carbohydrate + BCAA (CHO +BCAA)- 1 teaspoon BCAA powder (containing 2.5g leucine, 1.25g isoleucine, and 1.25g valine), mixed with 500ml a 6% CHO sports drink (Powerade Ion 4 ®- Orange, containing glucose, potassium, sodium, and vitamins B12, B3, and B6); and Placebo (PL): 500ml of a zero calorie sports drink (Powerade Ion 4 ZERO ®- Orange, containing potassium, sodium, and vitamins B12, B3, and B6). Ten minutes following ingestion, subjects performed a time cycle to exhaustion at 80% of their previously determined VO2 max, during which heart rate was
recorded continuously and ratings of perceived exertion were measured every 3 minutes using the validated Borg 1-10 RPE scale. It was hypothesized that 1.) Subjects receiving CHO + BCAA would significantly improve time to exhaustion when compared to BCAA and placebo, and 2.) Subjects receiving CHO + BCAA would demonstrate significantly lower average RPE scores during exercise when compared to BCAA and placebo.

Contrary to the hypotheses, the results revealed that cycling time to exhaustion was not significantly different between trials, $F(2,10) = .224, p > 0.05$. Average ratings of perceived exertion were also not significantly different between trials, $F(2,10) = 4.026, p = 0.052$.

INDEX WORDS: Carbohydrate, Branched-chain amino acids, Maximal oxygen uptake, Time to exhaustion
THE EFFECT OF A BCAA SUPPLEMENT WITH AND WITHOUT CHO ON PERFORMANCE IN COMPETITIVE CYCLISTS

by

ALAIN MILLS

B.S., Georgia Southern University, 2010

A Thesis Submitted to the Graduate Faculty of Georgia Southern University in Partial Fulfillment of the Requirements for the Degree

MASTER OF SCIENCE

STATESBORO, GEORGIA

2012
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Electronic Version Approved:
May 2012
DEDICATION

I would like to dedicate my thesis to my friends, family, coworkers, and professors who inspired and supported me throughout graduate school.

Special thanks to:

The GSU Cycling Club

Dr. Riggs

Dr. Rossi

Dr. Mcmillan

Dr. Joyner

Dr. Langdon

Dr. Romanchik

Alan & The CHHS Dean’s Office

Beth & Jody

Mom, Dad, Steve, Marissa & Lindsey
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CHAPTER 1
INTRODUCTION

Nutritional interventions prior to training and competition can greatly influence performance in both aerobic and anaerobic sports (Hoffman et al., 2009). This may help explain the rapid growth in use of pre-exercise supplements in recent years. The ability of a supplement to enhance performance is particularly useful to athletes who participate in prolonged activities on consecutive days that result in depleted muscle and liver glycogen (Skillen et al., 2008). Carbohydrate (CHO) in combination with protein as a pre-exercise, during exercise, and post-exercise supplement and/or ergogenic aid has been thoroughly investigated in recent research. While research first focused more on CHO+protein supplements for resistance training and power sport, attention has recently been turned to the potential benefits on endurance activities (Berardi, Noreen, & Lemon, 2008; Cepero, 2009; Ferguson-Stegall et al., 2010; Martinez-Lagunas, 2010). In particular, the branched-chain amino acids (BCAAs) have been investigated on exercise performance because of their potential in delaying the onset of central fatigue (Greer, White, Arguello, & Haymes, 2011).

Resistance exercise causes a number of metabolic changes in the body, stimulating both protein synthesis and degradation (Rasmussen, Tipton, Miller, Wolf, & Wolfe, 2000). It has been well documented that protein ingestion in combination with CHO before, during, or after resistance exercise can attenuate protein synthesis and keep the body in positive nitrogen balance, favoring muscle growth. (Beelen et al., 2008). The use of a CHO + protein supplement has been shown to decrease cortisol and other catabolic hormones in the body after resistance training (Beelen, et al., 2008; Bird, Tarpenning, & Marino, 2006). Consuming a BCAA drink
during the exercise recovery period may provide the muscle with an anabolic environment by stimulating insulin secretion and increasing the testosterone/cortisol ratio (Hsu et al., 2011). It has also been documented that BCAA consumption can help improve markers of cardiorespiratory fitness, such as VO₂ max (Matsumoto, Koba, Hamada, Tsujimoto, & Mitsuzono, 2009).

Protein in combination with CHO has also shown to enhance endurance performance in a number of ways. CHO ingestion prior to endurance exercise can improve performance by slowing the decline in blood glucose concentration (Byars, Greenwood, Greenwood, & Simpson, 2006). The addition of protein to a CHO supplement has been reported to potentiate the plasma insulin response of the supplement following a fast or prolonged aerobic exercise (Ivy, Res, Sprague, & Widzer, 2003). Berardi (2008) found that cyclists receiving protein + CHO improved time trial performance and felt less fatigued compared to those subjects receiving CHO alone. In addition, Cepero (2009) found that a CHO + protein supplement compared to CHO alone enhanced muscle recovery by decreasing lactic acid and increasing serum insulin levels. Even when matched for total calories, Niles and colleagues (2001) demonstrated that a CHO + protein drink resulted in greater performance gains in cycling activity than a supplement containing only CHO.

The use of pre-exercise supplements has become an increasingly popular practice among recreational and competitive athletes (Walsh, Gonzalez, Ratamess, Kang, & Hoffman, 2010). Drinks containing caffeine, CHO, and (BCAAs) are particularly common in endurance athletes. In addition, it is widely accepted that the ingestion of CHO during exercise can increase performance when the exercise duration is longer than 45 minutes (Jeukendrup, Brouns, Wagenmakers, & Saris, 1997).
**Purpose Statement**

The purpose of the current study was to examine the effect of a BCAA supplement with and without CHO on performance in competitive cyclists.

**Hypotheses**

1.) Subjects receiving CHO + BCAA will significantly improve time to exhaustion when compared to BCAA and placebo, and 2.) Subjects receiving CHO + BCAA will demonstrate significantly lower average RPE scores during exercise when compared to BCAA and placebo.

**Rationale**

Past research has shown that protein and CHO supplements positively influence endurance performance. However, a majority of the studies involved a glycogen depletion and resting period prior to supplementation (Berardi, et al., 2008; Ferguson-Stegall, et al., 2010; Howarth, Moreau, Phillips, & Gibala, 2009; Ivy, et al., 2003). As tapering periods are common during training, this method of testing may not properly represent an athlete’s physical state prior to competition. A number of research groups have examined whether BCAA supplementation might have a beneficial effect on endurance performance, but the results were inconsistent (Shimomura, Murakami, Nakai, Nagasaki, & Harris, 2004). To date, few research studies have conducted an experimental trial using BCAA’s + CHO in a pre-exercise supplement without a glycogen depletion phase. This study will seek to determine if taking a BCAA+CHO supplement prior to exercise will improve endurance performance in recreationally trained cyclists.
Central vs. Peripheral Fatigue

The onset of fatigue during endurance exercise is dependent on many factors. Physical fatigue, defined as the inability to maintain power output, can be either central or peripheral in origin. Central fatigue refers to an activity-induced inability to fully activate a muscle voluntarily, whereas peripheral fatigue implies that the ability of the muscle to produce force is reduced (Nordlund, 2003). Peripheral fatigue is caused mainly by depletion of muscle glycogen or phosphocreatine due to activity. Carbohydrate (CHO) administration prior to and during exercise may increase liver and muscle glycogen stores and provide a source of CHO during exercise. Maintaining or elevating body CHO stores may contribute to increased exercise performance, by delaying peripheral fatigue (Jentjens, Cale, Gutch, & Jeukendrup, 2003; Newsholme & Blomstrand, 2006).

There are many theories about the onset of fatigue in both central and peripheral aspects. The central governor theory proposes that a process in the brain regulates exercise capacity and the intensity of exercise to protect the heart from hypoxia (Noakes, Peltonen, & Rusko, 2001). The central governor is thought to limit exercise function by reducing muscle recruitment when exercise intensity is too high, and when muscle recruitment is lowered, cardiovascular function is limited and fatigue is experienced. According to this model, fatigue is only experienced after the onset of heart fatigue or failure (Noakes, et al., 2001).

Central fatigue is demonstrated experimentally when the maximal effort that can be achieved voluntarily is less than that which can be achieved when the muscle is stimulated
directly by electrical stimulation of the motor nerve (Newsholme & Blomstrand, 2006). The central fatigue hypothesis is based on the premise that an increase in serotonin levels in the brain during exercise results in the perception of fatigue. Tryptophan, an essential amino acid, is the precursor to serotonin. Transport of tryptophan into the brain is dependent on the concentration of free tryptophan in the blood and the ability of free tryptophan to compete successfully with the branched chain amino acids (BCAAs) (leucine, isoleucine, and valine) for transport to the brain (Manore, 2009). Therefore, administration of BCAAs prior to exercise can potentially delay the onset of central fatigue.

**Hormones Involved in Exercise**

Both aerobic and anaerobic exercises alter the plasma levels of certain substrates and hormones. These hormonal changes during exercise stimulate the breakdown of stored energy in the body. The intensity, duration, and mode of the exercise, as well as the nutritional status of an individual will influence the hormonal response to exercise. Anabolic hormones stimulate growth while catabolic hormones stimulate breakdown in the muscle tissue. Catabolic hormones (Cortisol, epinephrine, norepinephrine, and glucagon) are increased during exercise and signal the body to produce glucose for energy so that exercise can continue. Cortisol, produced in the adrenal cortex, stimulates gluconeogenesis, and helps to mobilize free fatty acids and amino acids, including the BCAAs. Epinephrine and norepinephrine (catecholamines) work together to elevate blood glucose and free fatty acid levels by promoting the breakdown of glycogen in both the liver and muscle and activating lipolysis, and blocking cellular glucose uptake. Glucagon responds when blood glucose levels are low, and stimulates both gluconeogenesis and glycogenolysis (Manore, 2009).
In general, anabolic hormones are decreased in the blood due to exercise. However, when nutrients are provided before, during, and/or after exercise, this trend can be reduced. Insulin, an anabolic hormone, is decreased in the blood due to exercise. Decreased levels of serum insulin will inhibit protein synthesis and cause increased protein degradation in the skeletal muscle. Growth Hormone (GH) increases protein synthesis and possibly inhibits breakdown of protein in the skeletal muscle (Manore, 2009). Insulin-like growth factor I (IGF-I) is an anabolic hormone that stimulates growth in almost all tissues, and is responsible for many of the effects of GH (Blomstrand, Hassmen, Ek, Ekblom, & Newsholme, 1997). Testosterone, another anabolic hormone, increases protein synthesis, and stimulates muscle growth. Unless nutritional interventions are implemented, these anabolic hormones will decrease due to exercise (Manore, 2009).

**Pre Exercise Supplements**

Drinks containing caffeine, CHO, and BCAAs are particularly common in endurance athletes research supports that the ingestion of CHO during exercise can increase performance when the exercise duration is longer than 45 minutes (Jeukendrup, et al., 1997). The goals of a pre-exercise meal, or of any food or supplement consumed just before competition, are to promote additional glycogen synthesis, to supply the body with glucose for use during exercise, and to minimize fatigue during exercise. A meal consumed within 1 hour before exercise should be small, easy to digest, and familiar to the individual (Manore, 2009).

A study conducted in 2010 by Walsh and colleagues investigated the effect of a pre-exercise supplement containing BCAAs on subjective measure of energy, focus and fatigue. Participants included fifteen active college students (9 men and 6 women), with a mean age of 20.9 ± 1.0 years. Prior to the experiment, VO2 max was determined on each subject. One week
later, subjects returned to the lab 3-hours post-absorptive and were randomly assigned to receive either the supplement (SUP) or placebo (P). The supplement, a commercially marketed drink ‘Amino Impact®’, consisted of 26 g of powder containing 20.5 g of caffeine, taurine, & glucuronolactone, 7.9 g of leucine, isoleucine, valine, arginine, & glutamine, 5g of di-creatine citrate, and 2.5 g of β-alanine. This supplement was mixed with 500 ml of water, for a total of 40 calories. Subjects who took the placebo received 500 ml of water sweetened with 3 g of sucralose and colored with red food coloring. The placebo contained no calories. Ten minutes after receiving either SUP or PL, subjects performed a timed cycle at 70% of their VO2 max on a treadmill. Time to exhaustion was determined as the time that the subject could no longer maintain exercise intensity or had reached volitional exhaustion (Walsh, et al., 2010). A 10 cm visual analog scale was used to assess subjective feelings of focus, energy, and fatigue prior, during and after exercise. A week after the first experimental trial, subjects returned to the laboratory and completed the same exercise protocol after receiving the opposite pre-exercise drink. Time to exhaustion was found to be significantly greater (p=0.012) in those subjects who received SUP as compared to PL. Subjects consuming SUP also reported significantly greater focus (p=0.031), energy (p=0.016), and significantly less fatigue (p=0.005) prior to exercise. Significant differences between groups were found following 10 minutes of exercise for focus (p=0.026) and energy (p=0.0004), but not fatigue (p=0.123). No differences were found post-exercise for any of the subjective measures. These results suggest that the acute consumption of Amino Impact® can enhance time to exhaustion during moderate intensity endurance exercise. Additionally, subjective feelings of fatigue, energy and focus may be improved.

Caffeine use as an ergogenic aid is common in all types of athletes, particularly young adults. Since 2002, the popularity of commercially available energy drinks has increased
substantially (Candow, Kleisinger, Grenier, & Dorsch, 2009). A study done by Candow (2009) investigated the effects of a caffeinated, sugar free solution on a high-intensity run time to exhaustion in young adults. Seventeen physically active college students (9 men, 8 women) with a mean age of 21 ± 4 years were randomly assigned to a supplement with sugar free Red Bull or a decaffeinated, sugar free placebo (lemon-lime flavored soft drink, tonic water, lime juice). The sugar free Red bull contained 2 mg/kg of caffeine. Sixty minutes after consumption of the drink or placebo, each participant performed a run time to exhaustion at 80% VO2 max. RPE and blood lactate were measured throughout the ride. There were no differences in run time to exhaustion (Red Bull 12.6 ± 3.8 minutes, Placebo 11.8 ± 3.4 minutes), perceived exertion (Red bull 17.0 ± 2.0, placebo 16.6 ± 1.8), or blood lactate between groups (Candow, et al., 2009). A possible reason for not finding an effect from sugar-free Red Bull may be due to the lack of CHO in the treatment solution. When combined, CHO and caffeine containing solutions have been shown to improve endurance performance (Candow, et al., 2009).

Pre-exercise supplements have shown benefits on both endurance and strength athletes. Hoffman (2009) examined the effects of an over-the-counter high energy supplement on physical performance and subjective feelings during exercise. Twelve strength and power athletes (with a mean age of 21.1 ± 1.3 years) were randomly assigned to receive SUP (Redline Extreme- 120 ml) or placebo (PL) prior to exercise. Following consumption of SUP or PL, subjects completed a survey to assess energy, fatigue, alertness, and focus. Experimental protocol then began, which consisted of a two-minute quickness and reaction test and a 20 second Wingate Anaerobic Power test. Subjective feelings of energy (3.5 ± 0.5 vs. 3.1 ± 0.5) and focus (3.8 ± 0.5 vs. 3.3 ± 0.7) were significantly higher during SUP compared to PL. Reaction time was also significantly
higher in SUP than PL. Although supplement ingestion did not have a significant effect on anaerobic power performance, it helped enhance subjective feelings during exercise.

In 2006, Byars et. al. examined the effectiveness of a pre-exercise supplement on indices on maximal cardiorespiratory fitness. College students (12 female and 12 male, with a mean age of 20.25 ± 1.42 years) completed two VO₂ max tests on a motor-driven treadmill. Each subject was given a pre-exercise drink or placebo thirty minutes prior to the test. The supplement consisted of a 7.2% CHO- electrolyte concentration, and the placebo was citrus flavored water. During the test, heart rate and time were measured continuously while blood pressure and RPE were measured every three minutes. The most significant finding was that VO₂ max and time were significantly greater in the SUP group vs. the PL group (P<0.05), indicating that the drink enhanced some aspects of cardiorespiratory fitness. Overall, increases were observed in VO₂ max (15.5%) and time to exhaustion (8.7%)(Byars, et al., 2006).

**Protein + CHO Supplementation and Aerobic Exercise**

Many athletes or active individuals tend to train or compete after an overnight fast. Since the liver provides glucose to the body during rest, the liver’s glycogen reserves will be nearly depleted overnight (Chryssanthopoulos, Williams, Nowitz, & Bogdanis, 2004). A pre-exercise supplement or meal containing carbohydrate can help replenish liver glycogen and provide glucose to the blood to prevent hypoglycemia. Endurance exercise also stimulates the oxidation of protein, especially when blood glucose levels are low. This indicates that additional protein may need to be provided to help repair muscle and keep the body in positive nitrogen balance (Manore, 2009). Coingestion of protein with CHO can also augment glycogen synthesis during recovery, particularly if CHO intake is suboptimal (Howarth, et al., 2009).
Berardi et. al. (2008) conducted a study to determine if there would be a difference in cycling time trial performance between CHO only and CHO and protein supplements. Fifteen trained cyclists reported to the lab and completed a 60 minute time trial on a cycle ergometer at either 5 or 7% constant grade. Ten minutes following the cessation of the time trial, each participant consumed a 1 liter post-exercise supplement containing CHO (100% maltodextrin, 3 g of crystal light®, and 1 liter of water) or CHO + Protein (33% maltodextrin, 33% glucose, and 33% whey protein hydrolysates, 3 g of crystal light®, and 1 liter of water), and a second and third identical drink at 60 and 120 minutes post-exercise. Six hours following the trial, the subjects completed another identical 60 minute time trial. Expired gases we analyzed during minutes 10-15 and 40-45 of the time trial. Blood samples were collected immediately pre-exercise, at minutes 15 and 45 during exercise, and at 5 minutes post-exercise. Prior to each time trial, subjects completed a Profile of Mood States (POMS) questionnaire (McNair, 2011) and a muscle soreness scale. Both groups performed significantly better during the first time trial, however, the reduction in distance traveled and power output during the second time trial was significantly less in those receiving CHO+ Protein (-0.30 ± 0.19 km and -3.86 ± 2.44 W) as compared to CHO (-1.05 ± 0.16 km and -16.50 ± 2.39 W). No significant differences were found between groups in muscle soreness. Subjects who received CHO + protein reported significantly smaller increases in Fatigue Inertia (+3.29 ± 0.47) vs. CHO (+8.57 ± 2.29)(Berardi, et al., 2008). The addition of protein to a CHO post-exercise supplement helped cyclists to better maintain subsequent time trial performance and power output. Along with performance improvements, POMS data also revealed that the subjects who ingested CHO + protein felt less fatigued, indicating that the benefit of CHO+ protein supplementation can be a combination of psychological and physiological.
The addition of protein to a CHO supplement has been reported to potentiate the plasma insulin response of the supplement following a fast or after prolonged aerobic exercise (Ivy, et al., 2003). Ivy et. al. (2003) compared the effects of a CHO-protein supplement with a CHO supplement on endurance performance. Nine trained cyclists with a mean age of 27.3 ± 1.3 years performed three experimental trials separated by seven days. Exercise protocol required each subject to cycle to fatigue on an ergometer. Subjects came to the lab after a 12 hour fast and were provided one of two supplements or a placebo in a double-blind, randomized, crossover design. All drinks contained the same amount of electrolytes and vitamins. The placebo contained only aspartame to add sweetness, but not calories. The CHO drink contained 7.75 g of CHO and 0.3 g of fat, while the CHO-PRO drink contained 7.75 g of CHO, 0.3 g of fat, and 1.96 g of protein. Supplements were provided in volumes of 200 ml immediately prior to exercise, and every 20 minutes thereafter until the exercise intensity was increased to 85% VO₂ max. Time to exhaustion was significantly increased in subjects receiving the CHO supplement when compared to placebo (CHO 19.7 ± 4.6 min vs. Placebo 12.7 ± 3.1 min). Furthermore, the CHO-PRO supplement resulted in a significant increase in time to exhaustion when compared to the CHO treatment (CHO-PRO 26.9 ± 4.5 min, p <0.05). Supplementation significantly increased plasma insulin, however, no differences were found in insulin response between the CHO and CHO- PRO supplements during exercise (Ivy, et al., 2003). The results of this study support the theory that adding protein to a CHO supplement can further delay fatigue in endurance exercise, but the mechanism for this improvement is unclear. It was hypothesized that the addition of protein would help to spare muscle glycogen, but the insulin responses between the supplements were not significantly different. A possible explanation for the difference in performance could
be due to the BCAAs in the CHO-PRO supplement delaying central fatigue, but further research is needed to confirm this theory (Saunders, Kane, & Todd, 2004).

The addition of extra protein calories to CHO beverages is practically important if it produces additional performance or recovery benefits (Saunders, et al., 2004). A study done in by Saunders et al. (2004) designed to address the following research questions: 1) Is cycling time to fatigue improved when subjects consume a CHO + P beverage versus an isocarbohydrate (but not isocaloric) CHO beverage? And 2) Does the consumption of a CHO + P beverage attenuate muscular damage compared with an isocarbohydrate (but not isocaloric) CHO beverage after exhaustive cycle ergometry (Saunders, et al., 2004)? Fifteen male trained cyclists with a mean age of 20.9 ± 3.3 completed this randomized, double-blinded study. Each subject performed two prolonged bouts of cycling to fatigue. In the first ride, subjects rode at 75% of their VO2 max at a self-selected cadence of >50 rpm until they were unable to maintain this minimum cadence for 30 seconds. Twelve to 15 hours after the initial exhaustive bout, each subject returned and repeated the exercise bout under the same conditions, except at a higher intensity (85% VO2 max) (Saunders, et al., 2004). Subjects consumed 1.8 ml/kg of preconstituted treatment fluid (CHO +P or CHO) every 15 minutes of exercise. The CHO drink had a 4:1 CHO/PRO ratio, such that the beverage had a CHO content of 26 g of CHO (~7.3 %) and a 6.5 g of whey protein (1.8%) per 355 ml of water. The CHO beverage was identically matched in CHO content with the CHO +P beverage, but lacked the protein calories (Saunders, et al., 2004). Metabolic measurements, RPE, blood glucose, and lactic acid measurements were taken every 30 minutes during exercise. Statistical analysis revealed that in the first ride, subjects rode 29% longer (p<0.05) when consuming the CHO+P beverage (106.3 ± 45.2 min) than the CHO beverage (82.3 ± 32.6 min). In the second ride, subjects performed 40% longer when consuming the
CHO+P beverage (43.6 ± 12.5) than when consuming the CHO beverage (31.2 ± 8.7 min). Peak post exercise plasma CPK levels, indicative of muscle damage, were 83% lower after the CHO+P trial (216.3 ± 122.0 U L) than the CHO trial (1318 ± 1935 U L). Adding protein to a CHO only recovery drink helped to increase performance as well as potentially enhance recovery. However, the differences found between the two beverages in this case could be due to the addition of calories in the CHO+P beverage.

It is possible that the increases in endurance performance seen with the addition of protein to a CHO supplement could be due to the increase in calories in a protein containing beverage. Therefore, Niles and colleges (2001) compared the effects of a CHO-PRO drink versus an isocaloric drink containing only CHO on the recovery process following CHO depleting exercise. Ten male runners (Mean age, 27 ± 2.5 years,  Mean VO2 max, 59 ± 5.5 ml/kg/min) participated in a randomized, double-blind, crossover study. On trial days, subjects came to the lab after a 12 hour overnight fast and completed a glycogen depleting run on a treadmill, at 80% VO2 max for the first 30 minutes and 70-75% VO2 max for the remainder of the run. Immediately post-exercise, each subject was given the appropriate supplement for that session. The CHO-PRO drink contained 112.0 g of dextrose and maltodextrin, and 40.7 g of protein consisting of milk and whey protein isolate mixture. The CHO drink contained 152.7 g of CHO, in the same ratio of dextrose and maltodextrin. Both supplements were ingested with water, for a total of 600 ml per dosage. The same serving of the supplement was taken 60 minutes after the initial dosage, and then a second running test was administered. After a 5 minute warm up participants performed a timed run to exhaustion on a treadmill. The criterion for termination of the test was when the individual voluntarily stopped (Madsen, MacLean, Kiens, & Christensen, 1996). The run time to exhaustion was longer during the CHO-PRO than CHO trial (p<0.05). At
90 minutes post-supplementation, the insulin levels were higher in the CHO-PRO than the CHO trial (p<0.05) (Madsen, et al., 1996). Plasma glucose levels were well maintained during both treatments. The addition of protein to a CHO supplement seemed to have enhanced the release of insulin in this case, even when the drinks caloric value was matched. The recovery process of muscle glycogen seemed to be accelerated when a drink which contains adequate amounts of both CHO and PRO is consumed compared to an isocaloric drink which only contains CHO (Madsen, et al., 1996).

In a similar study, Martinez-Lagunas et. Al. (2010) conducted a study to investigate the ability of PRO to reduce the need for CHO in a sports drink without reducing or possibly improving the efficacy of the drink relative to enhancing aerobic capacity (Martinez-Lagunas, 2010). Twelve trained cyclists (5 women and 7 men) completed 4 exercise trials, separated by 7 days, in a randomized manner. The exercise protocol involved cycling on a stationary bicycle at intensities that varied between 55 and 75% VO₂ max for 2.5 hours and then at 80% VO₂ max until fatigued (Martinez-Lagunas, 2010). One of four supplements was provided to each subject every 20 minutes during exercise. The supplements consisted of a 4.5% CHO plus 1.5% protein complex (CHO/PRO H), a 3% CHO plus 0.75% protein complex (CHO/PRO L), a 6% CHO supplement (CHO), or a placebo (PL). Time to fatigue was significantly increased in CHO, CHO/PRO H, and CHO/PRO L over placebo, with no significant differences between each supplement. Blood glucose, plasma insulin, and CHO oxidation were elevated above PL during the CHO, CHO/PRO H, and CHO/PRO L trials (Martinez-Lagunas, 2010). The authors concluded from these findings that partially substituting protein for CHO in a sports drink did not enhance performance; however, this change did not cause a decrease in performance.
Ferguson-Stegall et al., in 2010, conducted a similar study to assess the efficacy of a supplement containing a mixture of CHO (dextrose, maltodextrin, and fructose), yet a lower CHO content, and a moderate amount of protein, in extending time to exhaustion, and to determine if muscle damage would be reduced with the supplement containing protein compared to CHO alone despite containing 50% less CHO and 30% fewer calories (Ferguson-Stegall, et al., 2010). Fifteen trained endurance athletes, ranging in age from 20 to 40 years completed this study (8 men and 7 women). The study was conducted in a randomized, double-blind, crossover design, in which a 6% CHO beverage or a 3% CHO/1.2% protein was provided during exercise. The CHO beverage contained only dextrose, while the CHO-PRO beverage contained dextrose, maltodextrin, and fructose, and whey protein isolate. A volume of 275 ml of the selected beverage was provided at the beginning of exercise and every 20 minutes thereafter. Experimental protocol involved each participant to cycle at intensities alternating between 45 and 70% VO$_2$ max for 3 hours, after which the workload increased to ~74-85 % Vo$_2$ max until exhaustion. Time to exhaustion was significantly greater in CHO-PRO than CHO in subjects cycling at or below the ventilatory threshold. No significant overall treatment differences were found in plasma insulin, blood lactate levels, RPE, or HR was seen. Compared to a traditional CHO only supplement, a mixture of CHO plus a moderate amount of protein improves aerobic endurance at exercise intensities at or near the ventilatory threshold, despite containing lower total CHO and caloric content (Ferguson-Stegall, et al., 2010).

Thomas, Morris, & Stevenson (2009) hypothesized that there would be no difference between a CHO replacement (CR) and chocolate milk (CM) when matched for caloric content. Nine trained cyclists (Mean age- 25.4 ± 8.0 years) completed three experimental trials which consisted of a glycogen depletion trial, followed by a 4 hour rest and a timed cycle to exhaustion.
Subjects were provided a recovery drink within 60 seconds of the glycogen depletion trial; either CR (1 g CHO/kg) Fluid replacement (FR), or CM (volume calculated to provide a caloric content identical to CR- 62.9 ± 7.2 grams CHO 14.2g + 1.6g PRO, 9.2 ± 1.0g FAT). Following the recovery period, participants completed a cycle to exhaustion at 70% VO2 max. Cycle to exhaustion time was longer with CM than FR (51%) or CR (43%), indicating that the addition of protein helped enhance recovery following the glycogen depletion trial (Thomas, Morris, & Stevenson, 2009).

Cepero et al. (2009) set out to determine if endurance cycling performance, post-exercise recovery and muscle damage were altered when consuming a CHO only beverage versus a CHO and casein beverage. A total of 15 male cyclists with a mean age of 39.0 ± 9.8 years arrived to a lab after a 10 hour fast and completed a glycogen depletion cycling ride at 75% VO2 max and then consumed one of the test drinks in a randomized, double blind manner, and rested for 2 hours. The drinks consisted of CHO (70 g of CHO) or CHO + P (70g of CHO + 40g of casein). After the 2 hour recovery period, subjects completed a 20 km time trial as fast as possible. The results showed no significant differences in time taken in performing the 20 km ride when consuming the CHO beverage or the CHO+ P drink. Serum insulin concentrations were higher during recovery when CHO+P beverage was consumed (P<0.05). Glucagon and lactic acid levels increased more on the CHO than on the CHO+ P beverage (P<0.05) after the 20 km test. The CHO +P drink showed a different effect that the CHO only drink, indicating that adding protein to the drink enhanced recovery(Cepero, 2009).

During recovery periods after intense exercise, nutrient ingestion can alter the metabolic response and help with training adaptations. Howarth et al. (2009) set out to determine whether ingesting protein with CHO during recovery from prolonged exercise would increase mixed
skeletal muscle protein fractional synthetic rate and improve whole body protein balance compared with CHO alone. After consuming a standardized meal and performing a 2 hour bout of standardized cycling exercise, six trained men with a mean age of 22 ± 1 years ingested drinks that provided either 1.2 g CHO/kg/hr, 1.2g CHO/kg/hr, + 0.4 g of protein/kg/hr, or 1.6 g CHO/kg/hr. Beverages were ingested at a rate of 750/ml/hr for four hours. The CHO source of the drink was maltodextrin, and the protein source was hydrolyzed whey protein concentrate. Muscle biopsies and blood samples revealed that the fractional synthetic rate was higher (P<0.05) in PRO-CHO (0.09 ± 0.01%hr) vs. both L-CHO (0.07 ± 0.01%hr) and H-CHO (0.06 ± 0.01%hr). Whole body nitrogen balance was positive only during PRO-CHO; however, glycogen synthesis rate was not different between trials (Howarth, et al., 2009). The addition of protein to a CHO recovery beverage improved recovery in this case, even when the drink was matched for total calories. This indicates that protein after endurance exercise may be necessary for boosting fractional synthetic rate of protein, and improving nitrogen balance in the body, favoring growth.

Jentjens, et al. (2003) investigated the effects of ingesting differing amounts of glucose pre-exercise on the glucose and insulin responses during exercise in nine well-trained cyclists (29.6 ±2.4 years, VO2 max: 64.1 ± 2.1 ml/kg/min). Four exercise trials were completed by each subject in a randomized design. Subjects ingested a solution containing either 25g (LOW), 75g (MED), 400g (HIGH) of glucose or a glucose free placebo (PLAC) before performing 20 minutes of exercise at 72% VO2 max followed by a time trial at 80%. Plasma glucose concentration fell rapidly during steady state exercise in all trials except during PLAC. No difference in plasma glucose was found between the glucose trials at any time. However, there was no difference in time trial performance between the four trials. The ingestion of 0, 25, 75, 0r 200g of glucose pre-exercise did not affect time trial performance.
**Protein CHO Supplementation and Resistance Exercise**

It has been well documented that protein coingestion with CHO before, during, or after resistance exercise stimulates protein synthesis and keeps the body in positive nitrogen balance (Beelen, et al., 2008). In particular, the BCAAs (valine, leucine, and isoleucine) are used in skeletal muscle during exercise as an energy source. Resistance exercise usually results in post-exercise protein synthesis and breakdown, and rates of synthesis of must exceed the rates of protein breakdown in order for muscle to grow. When proper nutrients are provided, protein synthesis can exceed breakdown, creating an anabolic environment (Manore, 2009). When protein is ingested without CHO, the protein will be used to maintain blood glucose rather than protein synthesis. Research indicates that CHO and/or amino acid ingestion around the time of exercise modifies the acute biochemical events associated with resistance training, shifting the exercise-induced hormonal profile toward a profile more favorable for positive protein balance (Bird, et al., 2006).

Beelen et al. (2008) conducted a study to assess the surplus value of protein coingestion with CHO during resistance-type exercise on whole body protein balance and skeletal muscle protein synthesis rate under normal, practical conditions (Beelen, et al., 2008). Ten healthy male subjects participated in two trials in which they consumed either CHO (0.15g/kg/h CHO) or CHO with protein (0.15g/kg/h CHO with 0.15g/kg/hr whey protein) every 15 minutes during a 2 hour resistance exercise session. As expected, muscle biopsies revealed that protein coingestion lowered whole body protein breakdown rates (p=0.0006) compared to CHO only, and augmented protein oxidation and synthesis rates. Whole body net protein balance was negative in CHO, whereas a positive net balance was achieved after the CHO + PRO treatment.
Even in a fed state, consuming protein in combination with CHO before, during, and after exercise can help shift the body from a catabolic to an anabolic state, favoring muscle growth.

Bird et al. (2006) set out to examine the influence of nutritive interventions on acute biochemical responses to a single bout of resistance exercise in untrained young men. Thirty-two untrained young men (18 to 29 years old) participated in this study that involved randomized, double-blind, placebo controlled protocol. Subjects performed a single bout of heavy resistance exercise, during which they consumed one of four nutritive interventions (a 6% CHO solution, a 6-g EAA mixture, or a combined CHO+EAA Beverage, or Placebo: aspartame and citrus flavoring). Supplements were dissolved in water at a fluid volume of 8.5ml/kg body mass. Total volume of liquid was divided by 25 servings, allowing for 22.5 to 30.0ml to be ingested between each set of exercise. Blood samples were taken every 15 minutes throughout the exercise bout. Exercises included leg press, leg curl, leg extension, shoulder press, lat pull down, bench press, barbell bicep curl, and supine tricep extension. No significant change in glucose or insulin was observed for placebo, while CHO and CHO+ EAA resulted in significantly increased glucose and insulin concentrations. EAA resulted in significant postexercise increases in insulin only. The placebo group exhibited a significant increase in cortisol, while the treatment groups displayed no significant change in cortisol during the exercise bout, with no between-group differences (Bird, et al., 2006). CHO and/or EAA administration during a bout of resistance exercise suppresses the exercise-induced cortisol response, allowing for insulin release.

Koopman et al. (2005) designed an experiment to determine post exercise muscle protein synthesis and whole body protein balance following the combined ingestion of CHO with or
without protein and/or free leucine. Eight male subjects with a mean age of 22.3 ± 0.9 years were randomly assigned to receive one of three drinks after a 45 minute bout of resistance exercise. The drinks consisted of: CHO (CHO), CHO plus protein (CHO + PRO) or CHO plus protein and free leucine (CHO+PRO+Leu). The exercise protocol consisted of 16 sets of 8 repetitions on leg press and leg extension machines at 80% of subject’s 1 repetition maximum with two minute rest intervals(Koopman et al., 2005). Subjects then rested and received repeated boluses of the test drinks. A primed continuous infusion of phenylalanine was used to determine fractional synthetic rate of protein. Subjects receving CHO+PRO+Leu demonstrated a higher insulin response, lower protein oxidation rates, and greater whole body protein balance rates compared to CHO and CHO+PRO (all p<0.05). Protein balance was negative during the CHO trial but positive during the CHO+PRO and CHO+PRO+leu trials (Koopman, et al., 2005).

**BCAA Supplementation and Exercise**

Matsumoto et al. (2009) conducted a study involving eight trained male cyclists with a mean age of 21 ± 2 years. The purpose of the study was to determine the effect of BCAA supplementation before an incremental loading exercise test following 7-day supplementation on the lactate threshold. Subjects received either a BCAA drink (0.4% BCAA, 4% CHO; 1500 ml/day) or placebo (isocaloric, containing dextrin) for six days. An incremental loading exercise test on a cycle ergometer was performed on the 7th day. A total of 500ml of the test drink was given 15 minutes prior to the test. The VO2 max and workload levels at the lactate threshold were higher in the BCAA trial than that in the placebo trial (VO2: 29.8±6.8 vs. 26.4±5.4 mL/kg/min; workload: 175±42 vs. 165±38 W, p<0.05, respectively). The VO2max in the BCAA trial was higher than that in the placebo trial (47.1±5.7 vs. 45.2±5.0 mL/kg/min, p<0.05)
(Matsumoto, et al., 2009). These results indicate that VO₂ max and workload levels at the lactate threshold can potentially be improved by BCAA supplementation.

A study conducted in 2011 by Hsu et al. examined the effects of a BCAA drink on biochemical responses and psychological conditions during recovery after a single bout of exhaustive exercise (Hsu, et al., 2011). Fourteen physically active male college students (age 23.4 ± 0.8) participated in this randomized, double-blind, placebo-controlled crossover trial. Subjects were assigned to ingest 2 different test drinks, a BCAA drink (0.5 g valine, 1.0 g leucine, 0.5 g isoleucine, 0.5 g arginine, 12.1 g CHO, 100 ml water), or placebo 5 minutes after an exhaustive bout of exercise on a treadmill. Blood samples were collected before exercise, and at 10, 20, 40, 60, and 120 min during the recovery period. The levels of glucose and insulin were significantly higher in the BCAA trial as compared to those in the placebo trial at the 40 and 60 minute recovery points. The testosterone-to-cortisol ratio at the 120 minute recovery point was significantly higher in the BCAA trial as compared to that of the placebo (Hsu, et al., 2011). These results suggest that consuming a BCAA drink during exercise recovery period may provide the muscle with an anabolic environment by stimulating insulin secretion and increasing T/C ratio.

Tipton et al. (2001) recruited six recreationally active volunteers (3 females, 3 males) to determine whether an oral amino acid-CHO solution (EAC) would be a more effective stimulator of muscle protein anabolism if given immediately before or immediately after a resistance exercise bout (Tipton et al., 2001). Subjects consumed an amino acid CHO drink (6g of essential amino acids, 36g sucrose, 500ml of water) either immediately before (PRE) or immediately after (POST) an intense leg-exercise bout; with blood samples taken before, during, and after the
exercises were performed. Muscle biopsies from the *vastus lateralis* were also taken before and after the exercise bout. Amino acid delivery to the leg was increased during exercise and for 2 hours post-exercise in both trials. However, amino acid delivery was significantly higher in PRE than POST during the exercise bout and 1 hour post exercise (P<0.05). Total phenylalanine uptake across the leg was greater during PRE than POST (P=0.0002)(Tipton, et al., 2001). Consuming an amino acid CHO drink before or after exercise stimulates protein synthesis, but consuming this drink before exercise may result in greater muscle anabolism.

Blomstrand, et al. (1997), examined the effect of a BCAA solution on perceived exertion during cycling at 70% VO₂ max. Seven trained male subjects (25± 2.8 years, VO₂ max 4.70 ± 0.27 L min) completed the randomized, double-blinded study. After an initial max test, subject completed two experimental trials in the morning after they had performed exhaustive exercise the night before to deplete muscle glycogen. Subjects came to the lab in a 12 hour fast and were given a a BCAA solution or placebo every 20 mins during exercise. Exercise protocol involved a 60 min ride at 70% VO₂ max followed by another 20 minutes of maximal exercise. Every 10 minutes during exercise subjects rated their perceived exertion and mental fatigue on two different Borg scales. The RPE scale was used to measure perceived performance while the CR-10 scale was developed to measure perceived stress, discomfort, and annoyance (Borg 1990). Physical performance was measured as the amount of work done during the last 20 minutes of exercise at maximum intensity. The BCAA drink was individualized to provide 80mg of BCAA’s per kilogram of body weight. The solution was 40% valine, 35% leucine, and 25% isoleucine. No differences were found for cardiovascular data or physical performance.
However, during BCAA, subject’s ratings of perceived exertion were 5% lower, and ratings of mental fatigue were 15% lower than during placebo (Blomstrand, et al., 1997).

Greer, et al. (2011) found similar results when supplementing BCAA’s to nine untrained (21.6± 3.2 years, VO₂ max 36.3 ± 2.2 ml/kg/min) male subjects. In a double-blind, crossover design, subjects received a BCAA drink, an isocaloric CHO drink, or a placebo before and during a 90 minute cycling bout at 55% VO₂ max followed by 15 minute time trials. RPE and metabolic measurements were taken every 15 minutes during the cycling bout. Subjects maintained detailed dietary intake records for 3 days prior to, and on the day of each experimental trial. During the trial subjects were in a four hour fast. One serving of the BCAA drink contained 4.8g isoleucine, 12.2g leucine, and 7.3g of valine. Beverages were administered 5 minutes before exercise began, and at the 60-minute mark during the ride, for a total of 200 calories provided over the two time points. Subjects receiving CHO covered a greater cycling distance than placebo during the time trial (P<0.05), however, there was no difference between the BCAA and placebo trials (P>0.05). RPE was reduced during the BCAA trial when compared to placebo at two different time points. Authors concluded that although BCAA supplementation did increase blood concentrations of BCAA’s, it did not influence aerobic performance. BCAA supplementation did however attenuate RPE during the trial (Greer, et al., 2011).

Mikulski, et al. (2002) examined whether BCAA supplementation influences psychomotor performance, determined by the multiple choice reaction time (MRT), during short-term exercise. Sixteen physically active subjects (age 22.4 ±0.5 years, VO₂ max 53.6 ±16 ml/kg/min) performed two double blinded experimental trials, in which they consumed pudding with out without BCAA (7g) before performing a maximal exercise test. MRT was measured
before exercise, during the last two minutes of each exercise load, and at the 2\textsuperscript{nd}, 5\textsuperscript{th}, and 10\textsuperscript{th} minute of the recovery period. There was no effect of BCAA ingestion on the maximal workload achieved during the test (placebo: 331±6, BCAA: 334±8 W), the subjects VO\textsubscript{2} max (placebo: 50.2±1.7, BCAA: 48.9±1.1 ml/kg/min) or maximal HR (placebo: 195±2, BCAA: 193 ±2 bpm). BCAA, however, shortened MRT at rest in comparison with placebo (277±9 vs. 296±10 ms, p<0.05). Authors concluded that supplementation with BCAA improves psychomotor performance at rest and delayed its decrement at high exercise intensities.

Madsen, et al. (1996) examined the effects of glucose, glucose plus BCAAs, or a placebo on bike performance over 100km. In a double blind, randomized design, nine well trained subjects (29 ± 1.1 year, VO\textsubscript{2} max 63.1 ± 1.5 ml/kg/min) received 1) glucose, 2) glucose plus BCAA, or 3) a non-caloric placebo. During the trials, the subjects were required to complete three 100-km bicycle experiments separated by 7 days. Subjects were instructed to complete the 100 km trial as fast as possible, and throughout exercise, expired gas samples, heart rate, and blood samples were obtained at minutes 10, 30, 60, 90, 120, 150, and at the termination of exercise. No significant differences were observed among trials in performance time (p>0.05). In the glucose +BCAA trial, plasma BCAA levels increased after exercise. The respiratory exchange ratio was similar in the three trials during the first 90 min of exercise; thereafter it tended to drop more in the placebo trial than the CHO and CHO+BCAA trials. These data suggested that glucose nor glucose +BCAA enhanced performance during a 100 km cycling bout.

Watson et al. (2004) examined the effect of acute BCAA supplementation on prolonged exercise capacity in a warm environment. Eight healthy male subjects were recruited (28.5 ± 8.2
years; VO$_2$ max 4.14 ±0.48 l/min), and completed two randomized, double blinded experimental trials in a crossover design separated by 7 days. Subjects were instructed to record dietary intake and physical activity during the 2 days before the first trial, and to replicate this in the 2 days prior to the second trial. Subjects cycled in a warm environment to exhaustion at 50% of VO$_2$ max. Four 250 ml BCAA solution or placebo was administered at 30 minute intervals for 2 hours prior to exercise, and during exercise, an additional 150 ml was consumed every 15 minutes throughout exercise. A significant reduction in the plasma concentration ratio of free tryptophan to BCAA was observed during the BCAA trial when compared to placebo (P<0.001). Blood glucose and lactate concentrations were not different between trials (both p>0.05), and BCAA ingestion also had no effect on time to exhaustion (placebo 103.9±26.9 min; BCAA 110.0±29.2 min; p=0.129), indicating that ingestion of a BCAA solution prior to, and during, prolonged exercise, did not influence exercise capacity in a warm environment.
CHAPTER 3
METHODOLOGY

The purpose of this study was to examine the effect of a BCAA supplement with and without carbohydrate (CHO) on performance in competitive cyclists.

Subjects
Six male participants were recruited from the Georgia Southern University cycling club. Subjects were college students between the ages of 19 and 22 years old. The study was approved by the Institutional Review Board at Georgia Southern University, and each participant was informed of the possible participation risks before completing a written informed consent. Subjects were given a Physical Activity Readiness (PAR-Q) questionnaire which assesses an individual’s readiness for participation in exercise training programs. Subjects also completed a medical history questionnaire before participating in any procedures.

Anthropometric Measures
Subjects who agreed to participate in the study completed an assessment of height and body weight. All measurements were taken in the human performance lab on the same occasion. Body weight was measured using a Siltec® digital scale. Subjects were weighed in their workout clothes without shoes. Height was measured using a standard wall mounted stadiometer and recorded to the nearest centimeter. During measurement, the subject was asked to stand with his back against the stadiometer, with the scapulae and buttocks also in contact if possible, and
weight evenly distributed between both feet. The subject was also instructed to stand erect (stand up straight and look straight ahead) (Rockenbach, 2007).

**Exercise Protocol**

**Cardiorespiratory Fitness Test:** Each subject’s maximal oxygen uptake (VO\text{2}max) was determined on an electrically braked cycle ergometer (Lode, B.V., Groningen, Netherlands) using a graded exercise test (Cepero, 2009). Prior to test participation, participants were asked to adhere to the following pre-test instructions: 1) wear comfortable, loose fitting clothing, 2) drink plenty of fluids over the 24-hour period preceding the test, 3) avoid food, tobacco, alcohol, and caffeine for three hours prior to taking the test, 4) avoid exercise or strenuous physical activity the day of the test, and 5) get an adequate amount of sleep (6 to 8 hours) the night before the test (Byars, et al., 2006).

The ParvoMedics TrueOne® 2400 open circuit spirometry system was used for collection and analyses of expired gases continuously. The computerized metabolic measurement system used a Hans Rudolf 3814 (Kansas City, MO) pneumotachometer to measure ventilation. The TrueOne 2400 is a mixing chamber system that uses a paramagnetic oxygen analyzer (range 0–25%) and an infrared, single beam, single wave-length carbon dioxide analyzer (range 0–10%) (Crouter, Antczak, Hudak, DellaValle, & Haas, 2006). Prior to testing, the metabolic cart was calibrated according to the manufacturer’s directions. Calibration protocol involved a room air auto-calibration routine and a two-point gas calibration with a single gas tank (15.09% O2, 6.01% CO2). In addition, the flow meter was calibrated using a 3.000 l Hans Rudolf 5530 series syringe. This involved a five stroke calibration using different flow rates for each stroke.
(Crouter, et al., 2006). The ParvoMedics TrueOne® 2400 has been shown to provide accurate and reliable results for the measurement of gas exchange variables (Crouter, et al., 2006). The average temperature during the tests was 20°C and the average humidity was 54.0%.

Prior to testing, subjects were fitted with a mouthpiece, nose clip, and headgear. A polar heart rate strap with a sensor, interfaced with the ParvoMedics system, was also fitted around the subject’s chest so heart rate could be monitored continuously during the trial. After a 2 minute warm-up at 100 watts, the watts increased by 25 W every minute until volitional exhaustion was reached (Cepero, 2009). Volitional exhaustion was reached when the subjects’ respiratory exchange ratio was greater than 1.15, and pedal cadence could no longer be maintained.

**Performance Measurement:** Subjects performed a timed cycle to exhaustion 10 minutes after consuming one of three randomly assigned beverages (Walsh, et al., 2010). All preparation procedures and calibrations prior to the cycle to exhaustion were identical to the procedures prior to the initial VO2 max test. Over a two minute period following a two-minute warmup at 100 watts, the resistance was increased in two increments to that which represented 80% of the subject's VO2 max. Heart rate was measured continuously using a polar heart rate monitor, interfaced with the ParvoMedics system (Martinez-Lagunas, 2010). Participants were instructed to stay seated throughout the trial. Pedal cadence was monitored constantly via the ParvoMedics system, and participants were given a warning when their cadence dropped by ≥ 10 r/min for more than 20 seconds. The second time this occurred, the trial was terminated and time to exhaustion was recorded (Thomas, et al., 2009). Ratings of perceived exertion were measured using the validated Borg 1-10 RPE (Borg, 1982) scale every 3 minutes (Thomas, et al., 2009) during each trial. Averages of the RPE values for each trial were used for data analysis.
**Supplement Protocols**

In a randomized, double blind, crossover design, each subject came to the lab in a 12 hour post absorptive state (Walsh, et al., 2010) on three separate occasions and consumed one of three pre-exercise supplements 10 minutes before performing the cycle to exhaustion (Walsh, et al., 2010). Each trial was separated by one week (7 days). The three experimental trials differed only in the beverage consumed. Subjects were assigned at random to 1 of 3 treatment orders to ensure than no trial had an advantage in regards to an order effect (Greer, et al., 2011). The liquid supplements were mixed by someone other than the principal investigator to ensure the researcher did not know which supplement was being consumed by the subject at that time. Subjects were aware that they are taking a pre-exercise supplement, but were not given the order of consumption until the completion of the study. Supplement compositions are detailed below.

**BCAA Group:** 1 teaspoon BCAA powder (Optimum Nutrition® Instantized BCAA 5000mg Powder containing 2.5g leucine, 1.25g isoleucine, and 1.25g valine), mixed with 500 ml flavored water (Powerade Ion 4 ZERO®- Orange, containing potassium, sodium, and vitamins B12, B3, and B6); 20 total calories

**Carbohydrate + BCAA (CHO +BCAA) Group:** 1 teaspoon BCAA powder (containing 2.5g leucine, 1.25g isoleucine, and 1.25g valine), mixed with 500ml a 6% CHO sports drink (Powerade Ion 4 ®- Orange, containing glucose, potassium, sodium, and vitamins B12, B3, and B6); 140 total calories

**Placebo (PL):** 500ml flavored water (Powerade Ion 4 ZERO®- Orange, containing potassium, sodium, and vitamins B12, B3, and B6); zero calories
Study Time Line

**Week 1:** An informational meeting was held with all subjects. All experimental procedures were explained in detail. Each subject completed their PAR-Q and medical history questionnaire, and then signed an informed consent form. The timeline was discussed and exercise times were set.

**Week 2:** Anthropometric measurements were taken on each subject by the researcher. Each subject reported to the laboratory and completed a VO²max test on a cycle ergometer (Parvo Medics' TrueOne® 2400).

**Weeks 3-8:** Each subject completed three experimental trials separated by at least one week. Subjects reported to the lab overnight after a 12 hour fast. After resting for ten minutes, subjects consumed their randomly assigned test drink. Ten minutes following ingestion, subjects performed a cycle to exhaustion at 80% VO²max. During the following experimental trials, subjects crossed over and consumed the other test drinks. Trials differed only in the beverage consumed.

**Statistical Analysis**

Cycling time to exhaustion data was analyzed using an ANOVA with repeated measures. Participant’s average measures of ratings of perceived exertion were analyzed using an ANOVA with repeated measures. A criterion alpha level of $p \leq 0.05$ was used to determine statistical significance. Contrasts were used to follow up a significant ANOVA interaction. Each subject’s time to exhaustion and average RPE scores were also examined.
CHAPTER 4

RESULTS

Subject Characteristics

Six male endurance trained cyclists completed the study. All of the subjects reported in a 12 hour fasted state prior to each experimental trial. Descriptive statistics are summarized in Table 1.

<table>
<thead>
<tr>
<th>Variable</th>
<th>Mean ± SD</th>
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<tbody>
<tr>
<td>Age (years)</td>
<td>20.50 ± 1.22</td>
</tr>
<tr>
<td>Weight (kg)</td>
<td>72.98 ± 6.78</td>
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<tr>
<td>Height (cm)</td>
<td>174.00 ± 4.20</td>
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<tr>
<td>VO₂ max (ml/kg/min)</td>
<td>59.5 ± 7.9</td>
</tr>
<tr>
<td>BMI (kg/m²)</td>
<td>22.8 ± 1.8</td>
</tr>
</tbody>
</table>

Time to Exhaustion

An ANOVA with repeated measures was used to analyze cycling time to exhaustion data. Mauchly’s test indicated that the assumption of sphericity had been violated (p < 0.05); therefore the Greenhouse Geisser adjustment was used. The results show that cycling time to exhaustion was not significantly different between trials, F(2,10) = .224, p > 0.05.
An ANOVA with repeated measures was used to analyze average ratings of perceived exertion data. Ratings of perceived exertion gradually increased during the ride. The results show that average ratings of perceived exertion were not significantly different between trials, $F(2,10) = 4.026$, $p = .052$. 

**Ratings of Perceived Exertion** 

![Bar chart showing mean time to exhaustion (min) with standard deviation bars for Placebo, BCAA, and BCAA+CHO conditions. The values are 13.21 ± 2.49, 13.3 ± 2.33, and 13.71 ± 2.93 respectively.}
**Single-Subject Analysis**

**Time to Exhaustion:**

Three out of the six subjects cycled the longest when receiving BCAA+CHO. Figures 3-8 show each subject’s time to exhaustion.
Figure 6

Time to Exhaustion: Subject 4

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Time (min)</th>
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<tbody>
<tr>
<td>Placebo</td>
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<tr>
<td>BCAA</td>
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<tr>
<td>BCAA+CHO</td>
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Figure 7

Time to Exhaustion: Subject 5

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<th>Treatment</th>
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<td>BCAA+CHO</td>
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Average Ratings of Perceived Exertion:

Figures 9-14 show the each subject’s average ratings of perceived exertion during each ride. There was a trend toward a lower RPE during BCAA, however this trend was not significant (p = .052).
Figure 12

Subject 4: Average RPE

<table>
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Figure 13

Subject 5: Average RPE

<table>
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Figure 14

Subject 6: Average RPE

<table>
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<tr>
<td>BCAA+CHO</td>
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CHAPTER 5

DISCUSSION

The purpose of this study was to examine the effect of a branched-chain amino acid (BCAA) supplement with and without carbohydrate (CHO) on performance in competitive cyclists. The researchers hypothesized that the addition of CHO would improve actual and perceived performance during a ride at 80% VO\textsubscript{2} max. BCAAs, which are metabolized in the muscle during aerobic exercise, have been theorized to delay central fatigue, while CHO is known to delay peripheral fatigue caused by low blood glucose levels. The main finding in the study was that the addition of CHO to a drink containing BCAAs did not alter time to exhaustion or ratings of perceived exertion during the ride. These results suggest that the ingestion of BCAA’s prior to high-intensity exercise did not improve performance or perceived performance. This finding is in contrast with some studies (Ferguson-Stegall, et al., 2010; Walsh, et al., 2010), but is in agreement with others (Blomstrand, et al., 1997; Jamurtas et al., 2011; Madsen, et al., 1996).

CHO availability during exercise, and muscle glycogen levels are major determinants of endurance performance. CHO administration becomes even more important when muscle glycogen levels are low at the onset of exercise because it can slow the decline in blood glucose concentration (Kerksick et al., 2008). Exercise time to exhaustion is known to be directly related to intramuscular glycogen content (Cepero, 2009), as ingesting CHO pre-exercise may increase liver or muscle glycogen stores and provide a source of CHO during exercise. Maintaining or elevating body CHO stores before or during exercise may contribute to increased exercise performance (Jentjens, et al., 2003). Many researchers have found improvements in time to
exhaustion following the administration of CHO prior to exercise (Martinez-Lagunas, 2010), however, the same effect was not found in this study. The present data suggest that supplementation of BCAA’s with or without CHO did not affect performance during high-intensity endurance exercise.

Past research supports that well trained individuals can maintain substantial energy production from fat oxidation (Madsen, et al., 1996). Subjects in the current study were highly trained (Mean VO₂ max = 59.5 ± 7.9 ml/kg/min) (Neiman, 1995). Endurance training results in a number of adaptations in both the cardiovascular and neuromuscular systems of the body. The more fit an individual, the more they utilize fat over CHO during aerobic exercise. This is due to the muscle adapting to using fat as fuel by gaining more fat storage, increasing the amount of mitochondria which have the capacity to metabolize lipids, and increasing the clearance and tolerance of lactate. These adaptations allow for the source of fuel to shift from CHO to fat earlier in an endurance exercise bout (Martinez-Lagunas, 2010).

Furthermore, training at or near the lactate threshold is an adequate training stimulus for improving the threshold (Londeree, 1997). High intensity training leads to an increase in both the number and activity of lactate transporters in skeletal muscle membranes, resulting in a greater ability to rapidly clear lactate. An improvement in the lactate threshold allows for an individual to exercise at a higher intensity for a longer period of time. Additionally, ingestion of CHO immediately prior or during exercise may blunt the stimulation of fat oxidative pathways by raising plasma insulin and glucose concentrations and lowering plasma free fatty acid (FFA) levels during exercise, thereby causing a marked shift in substrate oxidation (Civitarese, Hesselink, Russell, Ravussin, & Schrauwen, 2005).
Various studies have shown that CHO ingestion before or during prolonged exercise can delay fatigue and improve cycling performance (Martinez-Lagunas, 2010; Saunders, et al., 2004); which is accomplished by maintenance of blood glucose levels and a reduction in muscle glycogen utilization. These effects are most important during the end of exercise, as exhaustion is reached and glycogen is depleted. The body’s glycogen stores are relatively small, and may be depleted toward the end of exercise or after an overnight fast (Madsen, 1996). The type of CHO in Powerade® is glucose and maltodextrins. These are rapidly oxidized CHO’s that enter the blood as glucose at a quick rate (Frayn, 1983). These The higher percentage of VO₂ max (80%) used in this study made the length of exercise much shorter than other studies, with the longest ride lasting 18.34 minutes. It is possible that the CHO given in the CHO+BCAA drink did not influence the ride since it took so little time to reach exhaustion.

Although it is widely accepted that CHO given pre-exercise can enhance performance, other studies have produced conflicting results, perhaps because of difference in the designs of the studies. Jentjens, et al.(2003) investigated the effects of ingesting differing amounts of glucose pre-exercise on the glucose and insulin responses during exercise in nine well-trained cyclists (29.6 ±2.4 years, VO₂ max: 64.1 ± 2.1 ml/kg/min). Four exercise trials were completed by each subject in a randomized design. Subjects ingested a solution containing either 25g (LOW), 75g (MED), 200g (HIGH) of glucose or a glucose free placebo (PLAC) before performing 20 minutes of exercise at 72% VO₂ max followed by a time trial at 80%. Plasma glucose concentration fell rapidly during steady state exercise in all trials except during PLAC. No difference in plasma glucose was found between the glucose trials at any time. However, there was no difference in time trial performance between the four trials. The ingestion of 0, 25, 75, or 200g of glucose pre-exercise did not affect time trial performance. The finding that differing
amounts of glucose did not affect pre-exercise performance supported data from other studies (Alberici et al. 1993; Sherman et. al 1991). An important finding was that ingestion of CHO before exercise did not alter exercise performance compared to the ingestion of an energy free placebo, similar to the present study.

Martinez-Lagunas and colleagues (2010) conducted a similar study using both male and female trained cyclists. One of four supplements was provided to each subject every 20 minutes during exercise. The supplements included 1) a 4.5% CHO plus 1.5% protein complex (CHO/PRO H), 2) a 3% CHO plus 0.75% protein complex (CHO/PRO L), 3) a 6% CHO supplement (CHO), or 4) a placebo (PL). Time to fatigue was significantly increased in CHO, CHO/PRO H, and CHO/PRO L over placebo, with no significant differences between each supplement. These results suggest that CHO improved time to fatigue, despite varying amounts of protein. Although the supplement given in this study was lower in CHO than the 6% solution used in the present study, supplements were given every 20 minutes, resulting in higher total amount of CHO provided to each subject. The cycling protocol used in this study was also different than the 80% VO₂ max ride used in the present study. Participants began cycling at intensities between 55 and 75% VO₂ max for 2.5 hours, and then at 80% VO₂ max until fatigued. Although the same intensity was used during the time to fatigue measure, the 2.5 hour ride prior to this could have greatly affected the substrates available during exercise (Martinez-Lagunas, 2010).

Watson et al. (2004) examined the effect of acute BCAA supplementation on prolonged exercise capacity in a warm environment, and found no effect on exercise capacity, similar to the present study’s findings. Eight healthy male subjects were recruited (28.5 ± 8.2 years; VO₂ max
4.14 ±0.48 l/min), and completed two randomized, double blinded experimental trials in a
crossover design separated by 7 days. Subjects were instructed to record dietary intake and
physical activity during the 2 days before the first trial, and to replicate this in the 2 days prior to
the second trial. Subjects cycled in a warm environment to exhaustion at 50% of VO₂ max. Four
250 ml BCAA solution or placebo were administered at 30 minute intervals for 2 hours prior to
exercise, and during exercise, an additional 150 ml was consumed every 15 minutes throughout
exercise. A significant reduction in the plasma concentration ratio of free tryptophan to BCAA
was observed during the BCAA trial when compared to placebo (P<0.001). Blood glucose and
lactate concentrations were not different between trials (both p>0.05), and BCAA ingestion also
had no effect on time to exhaustion (placebo 103.9±26.9 min; BCAA 110.0±29.2 min; p=0.129),
indicating that ingestion of a BCAA solution prior to, and during, prolonged exercise, did not
influence exercise capacity in a warm environment.

Madsen, et al. (1996) also found no enhancement in performance when supplementing
glucose or glucose +BCAA when compared to placebo in cyclists. In a double blind, randomized
design, nine well trained subjects (29 ± 1.1 year, VO₂ max 63.1 ± 1.5 ml/kg/min) received 1)
glucose, 2) glucose plus BCAA, or 3) placebo. The glucose solution consisted of a 5% CHO
solution, and the same solution with 18g of BCAAs was used for the glucose plus BCAA
solution. The placebo contained no calories. Exercise training and food intake were controlled
for three days prior to the exercise trials. During the trials, the subjects were required to
complete three 100-km bicycle experiments separated by 7 days. Subjects were instructed to
complete the 100 km trial as fast as possible, and throughout exercise, expired gas samples, heart
rate, and blood samples were obtained at minutes 10, 30, 60, 90, 120, 150, and at the termination
of exercise. No significant differences were observed among trials in performance time (p>0.05).

In the glucose +BCAA trial, plasma BCAA levels increased after exercise. The respiratory exchange ratio was similar in the three trials during the first 90 min of exercise; thereafter it tended to drop more in the placebo trial than the CHO and CHO+BCAA trials. These data suggested that glucose nor glucose + BCAA enhanced performance during a 100 km cycling bout.

The protocol chosen to determine time to exhaustion in the present study was similar to other studies using similar performance tests, however, the cyclists responded differently during the experimental trials. Subjects were instructed that they would be given a warning when their cadence dropped by ≥ 10 r/min for more than 20 seconds. The second time this occurred, the trial would be terminated and time to exhaustion would be recorded. However, the subjects would slow down and then stop without responding to the warning, causing the trial to be terminated early. A possible reason for this is the higher percentage of VO₂ max (80%) used in this study, making it harder for subjects to regain cadence once it had dropped.

The researchers hypothesized that the addition of CHO would lower the subject’s ratings of perceived exertion during exercise. RPE scores gradually increased during the course of exercise. Average RPE was lower with BCAA; however, this finding was not significant (p=0.052). Several studies have suggested that BCAA may improve cognition, focus, mood and psychomotor function (Blomstrand, et al., 1997; Greer, et al., 2011; Hsu, et al., 2011; Walsh, et al., 2010) The results of this study suggested a decrease in perceived exertion with supplementation of BCAA, however the finding was not significant. Lower RPE scores during BCAA could be due to delayed central fatigue in the brain. Administration of BCAA’s during or
before exercise can serve to decrease the amount of tryptophan to enter the brain, therefore decreasing the amount of serotonin and delaying central fatigue.

A comparable study done by Blomstrand (1997) concluded with similar results. Seven trained male cyclists received either placebo or a solution containing BCAA’s before a 60 minute cycle bout at 70% VO2 max. RPE scores on the 12 degree (6-20) Borg scale were taken every ten minutes during the ride. Using the CR-10 scale (Blomstrand, et al., 1997), The subjects were also instructed to rate their perceived degree of mental fatigue every 10 minutes. Blood samples were collected every 20 minutes during the exercise and 5 minutes after the end of exercise. Although there was no difference in the physical performance between trials, the subjects rated their overall exertion as higher during the placebo trial than during the BCAA trial.

A more recent study conducted by Greer and colleagues (2011) set out to determine whether BCAA supplementation would affect aerobic performance, RPE, or substrate utilization, compared to an isocaloric CHO drink or a noncaloric placebo. Nine untrained subjects performed three 90 minutes rides at 55% VO2 max followed by 15 minute time trials. RPE and metabolic measurements were taken every 15 minutes during steady exercise. The CHO drink was a 6% solution (Gatorade®) and an isocaloric BCAA containing beverage was used. The BCAA beverage contained 4.8g of isoleucine, 12.2g of leucine, and 7.3g of valine. Beverages were administered 5 minutes before the initiation of exercise, and at the 60 minute mark. A significantly greater distance was traveled in the time trial during CHO than placebo (3.9 ± 0.4 km) (p<0.05). There was no difference between the BCAA and placebo trials. RPE was reduced at the 75 minute and 90 minute mark during the BCAA trial as compared with the placebo. The authors concluded that BCAA supplementation did not influence aerobic performance but did attenuate RPE when compared with the placebo (Greer, 2011).
Hsu and colleagues (2011) examined the effects of a post-exercise BCAA drink containing BCAA’s arginine, and CHO compared to a noncaloric placebo. After a single bout of exhaustive exercise on a treadmill, the levels of glucose and insulin were higher in the BCAA trial than placebo 40 and 60 minutes after exercise. Subjects were given a Profile of Mood States Assay to assess tension, depression, anger, vigor, fatigue, and confusion prior to exercise, and 120 minutes during the recovery period. The decrease in fatigue score at the 120 minute recovery point was significant only in the BCAA trial (Hsu, et al., 2011).

**Beverage Description**

All beverages contained the same amount of fluid, sodium, and potassium. The BCAA and BCAA+CHO drinks contained the same amount of BCAA’s (5g total: 2.5g leucine, 1.25g isoleucine, and 1.25g valine), but were not matched for total calories. Each supplement was sweetened with the same flavoring so that the subjects would not be aware of which drink they were consuming. Table 2 displays the nutrient composition of each supplement.

<table>
<thead>
<tr>
<th>Treatment</th>
<th>Placebo</th>
<th>BCAA</th>
<th>BCAA+CHO</th>
</tr>
</thead>
<tbody>
<tr>
<td>Grams CHO</td>
<td>0</td>
<td>0</td>
<td>30</td>
</tr>
<tr>
<td>Grams BCAA</td>
<td>0</td>
<td>5 *</td>
<td>5 *</td>
</tr>
<tr>
<td>Grams Fat</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>Sodium (mg)</td>
<td>200</td>
<td>200</td>
<td>200</td>
</tr>
<tr>
<td>Potassium (mg)</td>
<td>50</td>
<td>50</td>
<td>50</td>
</tr>
<tr>
<td>Total Milliliters</td>
<td>500</td>
<td>500</td>
<td>500</td>
</tr>
</tbody>
</table>
*2.5g leucine, 1.25g isoleucine, and 1.25g valine

A common method used in research when administering CHO is grams per kilogram of body weight (Beelen, et al., 2008; Saunders, et al., 2004). The current study, however, used a standardized 6% CHO solution, because the researchers wanted to examine the effect of a commonly ingested pre-exercise drink. The BCAA’s used in the supplements were a 2:1:1 ratio of leucine: isoleucine: valine, respectively. Although the most effective ratio of BCAA’s has not yet been determined in research, this is the most common ratio used because this is similar to that of animal protein. Leucine is the most potent amino acid among the BCAA’s for stimulating protein synthesis, and insulin release from the pancreas (Shimomua, 2004). Additional studies are needed to confirm if this ratio is the most effective. The five grams of BCAA’s given in the supplement was based on the recommended serving size of the supplement. Without blood analysis, it is not possible to determine whether this amount was sufficient in increasing the plasma concentration of BCAA’s, or influencing the ratio of free tryptophan to BCAA.

The traditional 6% CHO solution is the most widely researched pre-exercise supplement, and many studies have demonstrated improvements when using this 6% solution prior to exercise (Ivy, et al., 2003; Martinez-Lagunas, 2010). During endurance exercise, CHO is oxidized at about 1g/min (Jeukendrup, 2004); however, the high intensity of the exercise could have increased this oxidation. Although the BCAA+CHO beverage administered in the present
study provided a total of 30 grams of CHO, it is possible that a greater amount would have produced significant results.

Even though the experimental design was double-blinded, randomized, and placebo-controlled, this study still had limitations. It was up to the subjects to be in a fasted state, and refrain from exercise, caffeine, and nicotine at least 12 hours prior to measurement, and although each subject reported to be in a 12-hour post-absorptive state prior to each trial, diet was not controlled the days before the trials. The composition of the subject’s diets in the days prior to the experimental trials could have influenced liver and muscle glycogen levels. There is evidence that higher than normal pre-exercise muscle glycogen contents increase the time to exhaustion and performance in longer duration exercise (Bussau, 2002). If the subjects were following a low CHO diet prior to the experimental trial, their glycogen levels could be especially low, causing a decrease in endurance performance. In contrast, a high CHO diet prior the experimental trial could result in increased performance due to supersaturating the glycogen stores. Glycogen levels could also be especially low if subjects consumed a meal with inadequate calories the night before an experimental trial. The subjects were instructed to drink plenty of fluids over the 24-hour period preceding the test, avoid exercise of strenuous physical activity the day of the test, and to get and adequate amount of sleep (6 to 8 hours) the night before the test, however following these instructions was entirely up to the subjects. It is also possible that stress due to school and cycling team training could have had an influence on the test results on any given day. Another limitation of the present study was the fairly small sample size (n=6); for this reason, these findings cannot be generalized to the broader community. Although the treatment order was randomized, the selection of the subjects was non-randomized. This study
was limited to members of the Georgia Southern University cycling team, and for that reason the results may not be generalized to other populations.

Consuming a pre-exercise supplement containing BCAA’s with or without CHO did not appear to have an effect on performance or perceived performance in competitive cyclists. In the future, more research needs to be done to determine the proper ratio and amount of BCAA’s to provide in a supplement in order to produce an ergogenic effect. Future studies should have subjects follow a standardized diet in the day or days prior to experimental trials to eliminate any effect of diet. Blood analyses can help researchers examine parameters such as blood levels of BCAA’s, blood glucose and insulin levels, and blood lactate during exercise. Using a lower percentage of VO₂ max could allow the subjects to exercise for longer and possibly allow for the supplement to have a significant effect.

REFERENCES


Madsen, K., MacLean, D. A., Kiens, B., & Christensen, D. (1996). Effects of glucose, glucose plus branched-chain amino acids, or placebo on bike performance over 100 km. [Clinical Trial
Controlled Clinical Trial
Research Support, Non-U.S. Gov't]. *Journal of applied physiology, 81*(6), 2644-2650.


The Journal of experimental biology, 204(Pt 18), 3225-3234.


10.1186/1550-2783-7-14
APPENDIX A

PAR-Q & YOU

(A Questionnaire for People Aged 15 to 69)

Regular physical activity is fun and healthy, and increasingly more people are starting to become more active every day. Being more active is very safe for most people. However, some people should check with their doctor before they start becoming much more physically active.

If you plan to become much more physically active than you have been, start by answering the seven questions in the box below. If you are over the age of 69, the PAR-Q will tell you if you should check with your doctor before you start. If you are over 69 years of age, and you are not used to being very active, check with your doctor.

Common sense is your best guide when you answer these questions. Please read the questions carefully and answer each one honestly: check YES or NO.

<table>
<thead>
<tr>
<th>YES</th>
<th>NO</th>
</tr>
</thead>
<tbody>
<tr>
<td>1. Has your doctor ever said that you have a heart condition and that you should only do physical activity recommended by a doctor?</td>
<td></td>
</tr>
<tr>
<td>2. Do you feel pain in your chest when you do physical activity?</td>
<td></td>
</tr>
<tr>
<td>3. In the past month, have you had chest pain when you were not doing physical activity?</td>
<td></td>
</tr>
<tr>
<td>4. Do you lose your balance because of dizziness or do you ever lose consciousness?</td>
<td></td>
</tr>
<tr>
<td>5. Do you have a bone or joint problem (for example, back, knee or hip) that could be made worse by a change in your physical activity?</td>
<td></td>
</tr>
<tr>
<td>6. Is your doctor currently prescribing drugs (for example, water pills) for your blood pressure or heart condition?</td>
<td></td>
</tr>
<tr>
<td>7. Do you know of any other reason why you should not do physical activity?</td>
<td></td>
</tr>
</tbody>
</table>

If you answered YES to one or more questions:

- Talk with your doctor by phone or in person BEFORE you start becoming much more physically active or BEFORE you have a fitness appraisal. Tell your doctor about the PAR-Q and which questions you answered YES.
- You may be able to do any activity you want — as long as you start slowly and build up gradually. Or, you may need to restrict your activities to those which are safe for you. Talk with your doctor about the kind of activities you wish to participate in and follow his/her advice.
- Find out which community programs are safe and helpful for you.

If you answered NO to all questions, you can be reasonably sure that you can start becoming much more physically active — begin slowly and build up gradually. This is the safest and easiest way to go.

- Take part in a fitness appraisal — this is an excellent way to determine your basic fitness so that you can plan the best way for you to be active. It is also highly recommended that you have your blood pressure evaluated. If your reading is over 140/90, talk with your doctor before you start becoming much more physically active.

DELAY BECOMING MUCH MORE ACTIVE:

- If you are not feeling well because of a temporary illness such as a cold or a fever — wait until you feel better.
- If you are or may be pregnant — talk to your doctor before you start becoming more active.

PLEASE NOTE: If your health changes so that you may answer YES to any of the above questions, tell your fitness or health professional. Ask whether you should change your physical activity plan.

No changes permitted. You are encouraged to photocopy the PAR-Q but only if you use the entire form.

NOTE: If the PAR-Q is being given to a person before he or she participates in a physical activity program or fitness appraisal, this section may be used for legal or administrative purposes.

"I have read, understood and completed this questionnaire. Any questions I had were answered to my full satisfaction."

NAME: ____________________________
SIGNATURE OF PATIENT: ____________________________
DATE: ____________________________
SIGNATURE OF PARENT or GRANDPARENT (if participant under the age of majority): ____________________________
WITNESS: ____________________________

Note: This physical activity clearance is valid for a maximum of 12 months from the date it is completed and becomes invalid if your condition changes so that you would answer YES to any of the seven questions.
Code: ___________

Participants Name: ____________________________________
Today’s date: ___________________

Name of Physician: ___________________ Physician Phone: ______________________

Person to notify in case of emergency: ___________________ Phone: ______________________

**Article I. PLEASE CHECK ANY CONDITIONS THAT APPLY TO YOUR PAST OR PRESENT MEDICAL STATUS:**

- □ High blood pressure
- □ Heart murmur
- □ Heart attack
- □ Angina / chest pains
- □ Mitral valve prolapse (MVP)
- □ Heart valve prosthesis
- □ Pacemaker
- □ Congenital heart defects
- □ Rheumatic fever
- □ Artificial implants or grafts (joint prosthesis)
- □ Arthritis
- □ Stroke
- □ Glaucoma
- □ Seizures / convulsions
- □ Psychiatric treatment
- □ Difficult swallowing
- □ Diabetes
- □ Cancer
- □ Alcoholism
- □ Kidney disease
- □ HIV positive
- □ AIDS
- □ Ulcer
- □ Colitis
- □ Headaches
- □ Hepatitis
- □ Jaundice
- □ Prolonged bleeding
- □ Anemia
- □ Hemophilia
- □ Asthma
- □ Emphysema
- □ Tuberculosis
- □ Sinus infection
- □ Earaches
- □ Thyroid disease
- □ Venereal disease
- □ Recreational drug use

- □ Do you use tobacco? ........... If yes, how long?______

- □ I am or may be pregnant / nursing_______________

- □ Please list any known food allergies:
  ___________________________________________________________________________
  ___________________________________________________________________________
  ___________________________________________________________________________

- □ I have been under the care of a physician or have been hospitalized during the past two years. _________________________________________________________________

- □ I have a disease, condition or problem not listed that you should know about.
  ___________________________________________________________________________

- □ I am currently taking, or have recently taken the following medications (including antibiotics, cortisone, pain medication, birth control pills, antidepressants, sleeping pills).
  ___________________________________________________________________________
  ___________________________________________________________________________
I certify the above information is true and correct to the best of my knowledge.

_______________________________________________________         ______________
Signature of participant                                                                    Date

_____________________________________________________________           _______________
Signature of Investigator        Date
APPENDIX C
IRB INFORMED CONSENT

1. **Principal Investigators:**

   Alaine Mills, Graduate student, Department of Health and Kinesiology, 678-425-4331

   Amy Jo Riggs, RD, Ph.D., Assistant Professor, Department of Health and Kinesiology, 478-7753

2. **Purpose of the Study:** The purpose of this study is to determine the effect of pre-exercise drinks on a timed cycle to exhaustion in competitive cyclists. It is our hypothesis that a pre-exercise supplement containing the branched-chain amino acids plus carbohydrates will positively influence performance significantly more than a supplement containing only the branched-chain amino acids or a placebo containing no calories. Both branched chain amino acid and carbohydrate supplements have been researched on exercise performance, but never together in a pre-exercise supplement. The results of the study will be beneficial to athletes who are interested in improving performance by taking a supplement before competition.

3. **Procedures to be followed:** You will be asked to fill out an initial questionnaire to determine if you are eligible for the study. Participants with coronary heart disease, diabetes, and respiratory problems such as asthma will be excluded from the study. Your height, weight, and cardiorespiratory fitness will initially be tested on a stationary bicycle in the Human Performance Lab in Hanner Complex. You will then return to the lab on three separate occasions at least one week apart and consume one of three pre-exercise supplements 10 minutes before performing a cycle to exhaustion on a stationary bicycle. The supplements will include 1) **BCAA Group:** 1 teaspoon BCAA powder (Optimum Nutrition® Instantized BCAA 5000mg Powder containing 2.5g leucine, 1.25g isoleucine, and 1.25g valine), mixed with 500 ml flavored water (Powerade Ion 4 ZERO®- Orange, containing potassium, sodium, and vitamins B12, B3, and B6); 2) **Carbohydrate + BCAA (CHO + BCAA) Group:** 1 teaspoon BCAA powder (containing 2.5g leucine, 1.25g isoleucine, and 1.25g valine), mixed with 500ml a 6% CHO sports drink (Powerade Ion 4®- Orange, containing glucose, potassium, sodium, and vitamins B12, B3, and B6); and 3) **Placebo (PL):** 500ml flavored water.
(Powerade Ion 4 ZERO ®- Orange, containing potassium, sodium, and vitamins B12, B3, and B6). Each supplement will be given in a randomized order, and you will not be informed of what order you are consuming them. At the completion of the study, you will be informed of the order in which you consumed the supplements and any effect they may have had on your performance. You will be asked to rate your perceived exertion during each exercise bout. Prior to each trial you will also be asked to fill out a 24-hour dietary recall.

4. **Discomforts and Risks:** Risks associated with aerobic fitness testing are minimal in a healthy population, but include lightheadedness, fainting, muscle soreness, and the extremely rare risk of cardiovascular complication or death. Risks will be minimized through examination of medical health history forms to determine any health risks for aerobic fitness testing. Personnel administering tests are certified in CPR, AED use and First Aid.

5. **Benefits:** A pre-exercise supplement that is effective in increasing performance is beneficial for athletes. A liquid supplement is convenient for busy and active individuals. Decreasing fatigue during exercise can make exercise more enjoyable, and increasing the time to fatigue during exercise can help athletes improve performance and meet individual goals. At the completion of the study, you will find out the order of supplement you received during the different trials and how it affected your performance.

6. **Duration/Time:** Each session in the human performance lab will last up to two hours. The duration of the study will be about 3 months.

7. **Statement of Confidentiality:** All scientific and personal data collected on subjects for presentation purposes will be kept confidential and stored in a locked file drawer in Hollis 2121-A. This information will be available only to the principal investigators. Your identity will not be revealed in publications or presentations that result from this study so as to protect your privacy and confidentiality. All data will be reported as means and standard errors.

8. **Right to Ask Questions:** You have the right to ask questions and have those questions answered. If you have questions about this study, please contact Alaine Mills, graduate student, Department of Health and Kinesiology, 678-425-4331, amills13@georgiasouthern.edu or Dr. Amy Jo Riggs, RD, Ph.D., Assistant Professor, Department of Health and Kinesiology, 478-7753, ajriggs@georgiasouthern.edu. For questions concerning your rights as a research
participant, contact Office of Research Compliance IRB@georgiasouthern.edu or call (912) 478-0843.

9. **Compensation:** There is no compensation for participating in the present research project.

10. **Voluntary Participation:** Your participation in this study is entirely voluntary. If you decide to participate, you are free to withdraw your consent and to stop participating at any time without penalty or loss of benefits to which you are otherwise entitled.

11. **Penalty:** If you decide not to participate, you will not be penalized, and you will not lose any benefits or services to which you are otherwise entitled.

12. You must be 18 years of age or older to consent to participate in this research study. If you consent to participate in this research study and to the terms above, please sign your name and indicate the date below.

You will be given a copy of this consent form to keep for your records. This project has been reviewed and approved by the GSU Institutional Review Board under tracking number H12091.

**Title of Project:** The effect of pre-exercise drinks on a timed cycle to exhaustion in competitive cyclists.

**Principal Investigators:**

Alaine Mills, Graduate student, Department of Health and Kinesiology, 678-425-4331
Amy Jo Riggs, RD, Ph.D., Assistant Professor, Department of Health and Kinesiology, 478-7753

______________________________________  _____________________
Participant Signature     Date

I, the undersigned, verify that the above informed consent procedure has been followed.

______________________________________  _____________________
Investigator Signature     Date