THE EFFECTS OF BRANCHED CHAIN AMINO ACIDS ON SORENESS AND POWER RECOVERY FOLLOWING AN ECCENTRIC EXERCISE BOUT IN RESISTANCE TRAINED AND NON-RESISTANCE TRAINED MALES.

By

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CHAPTER I

INTRODUCTION

Introduction

Physical activity, athletic performance, and recovery from exercise are enhanced by optimal nutrition¹. However, there is a great deal of conflicting and confusing information available regarding these concepts². Of the macronutrients, adequate amounts of protein are necessary for muscle hypertrophy. It is well accepted that strength and power athletes have a protein requirement which may be twice that of sedentary individuals¹. The greater amount of protein needed by these athletes is thought to enhance the recovery and remodeling processes of muscle fibers that have been damaged or disrupted during resistance exercise³.

A single bout of exercise increases muscle protein synthesis⁴ and muscle protein breakdown resulting in an increased muscle protein turnover⁵. Muscle protein synthesis must be greater than muscle protein breakdown for there to be an increase in muscle mass⁵. Following an exercise bout, an increase in the transport of amino acids may contribute to the greater stimulation of protein synthesis⁶. Taking advantage of this increase in transportation of amino acids by ingesting amino acids before and/or after an exercise bout has been researched. The ingestion of oral essential amino acids results in a change from net muscle protein degradation to net muscle protein synthesis after heavy resistance exercise in humans⁷. Essential amino acids must be obtained from the diet while non-essential amino acids can be synthesized in the body². Therefore, we rely on dietary sources to obtain the essential amino acids necessary to promote growth, repair damaged cells and tissue, and for a variety of metabolic and hormonal activities.

Of the essential amino acids, the branched chain amino acids (BCAA) have gained much attention in regards to muscle recovery. Garlick et al, found protein synthesis was stimulated equally by an essential amino acid mixture and a BCAA mixture when administered to young rats, but not equally stimulated by the non-essential amino acid mixture. This demonstrates the BCAAs are the key amino acids which stimulate protein synthesis⁸.

The branched chain amino acids have also been found to decrease muscle soreness⁹ and increase muscle recovery¹⁰ from eccentric muscle damage. Muscle soreness was decreased after squat exercise with a single supplementation of only 5 g of BCAA in both females and males for several days following exercise¹⁰. Jackman et al. demonstrated that muscle soreness was reduced significantly in a period of three days with supplementation of 7.3 grams of BCAA taken 4 times a day in untrained males⁹.

A single meal of a protein mixture ingested post-exercise increases the rate of force and power restoration¹¹. When a supplement of whey protein hydrolysate was ingested following an eccentric exercise bout, the consumption of this single dose resulted in a more rapid recovery of muscle force-generating capacity¹². However, neither of the previous studies' participants were resistance trained.

Training status affects the post exercise stimulation of muscle protein synthesis by altering the extent and duration of this process. A trained body will synthesize protein in a shorter time period by returning to basal protein synthesis more rapidly than an untrained body¹³. It is well known additional protein is needed along with sufficient energy to support muscle growth^{1, 6, 14, 15}. However, the amount of protein needed to maintain muscle mass may be lower for individuals who resistance-train routinely due to more efficient protein utilization¹⁵. Other data suggest that the muscle protein synthesis response to resistance exercise becomes more "refined" with training such that available substrates are directed toward the synthesis of different protein fractions depending on the exercise stimulus¹³.

Statement of the Problem

It has been demonstrated that untrained individuals benefit from BCAA supplementation by a decrease in perceived muscle soreness as well as more rapid recovery of muscle force-generation. However, current evidence suggests trained individuals do not synthesize protein in the same manner or amount of time as untrained individuals^{7, 13}. Further research is warranted to examine whether resistance trained individuals will have similar results as untrained individuals to BCAA supplementation following an eccentric exercise bout.

Purpose of the Study

The purpose of the current study is to determine if BCAA ingestion following an acute bout of eccentric exercise can attenuate muscle soreness and improve recovery similarly between resistance trained and untrained men.

Definition of Terms

<u>Trained Individuals</u> – Persons involved in lower extremity resistance training at least two times a week for the previous 10 weeks¹⁶.

<u>Untrained Individuals</u> – subjects not partaking in resistance training in the previous 6-9 weeks⁹.

<u>1-Repetition Max (1-RM)</u> – Assessment of dynamic strength to determine how much weight an individual can lift for one repetition.

<u>Muscle Soreness</u> – perceived muscle soreness rated using a 100 mm Visual Analog Scale (VAS)

<u>Muscular Power</u> – Output produced during a single, maximum effort muscle action. <u>TENDO Weightlifting Analyzer V-207</u>– the TENDO unit consists of a transducer attached to a belt to be worn by participants which measures linear displacement and time. Both peak and mean power output can be measured using this device³. <u>Visual Analog Scale (VAS)</u> - consists of a 100-mm line with anchor points of "no soreness" on the left and "extremely sore" on the right.

Significance of the Study

To our knowledge, this is the first study to compare trained individuals to untrained individuals in acute muscle soreness and power recovery with BCAA supplementation following an eccentric exercise bout. This is significant because trained individuals have different needs than untrained individuals caused by training adaptations and many times are not aware of these differences. This information can be used by professionals working with athletes as well as professionals conducting future research.

Assumptions

- All subjects maintained their typical diet during their participation in the study and therefore protein intake remained steady throughout the experimental trial. A dietary food log was expected to be completed by all participants to ensure no extreme, abnormal dietary practices were participated in during the experimental trial.
- 2. All subjects in the trained groups will be at the same level of trained and the subjects in the untrained group will be at the same untrained level.
- 3. Subjects were able to differentiate muscle soreness from muscle fatigue.
- 4. Subjects were able to express muscle soreness on the VAS adequately.
- Subjects were honest about protein supplementation both in the past and during the experimental trial.
- 6. Subjects consumed their supplements at the scheduled times.

Limitations

- 1. Small sample sizes (≤ 10 subjects per group)
- 2. The inability to control the subject's dietary intake, especially protein, carbohydrates and total energy.
- The possibility individuals did partake in exercise during the three days of measurements following the eccentric exercise bout. Certain exercises or even common daily activities may affect the amount of perceived soreness and muscle function recovery.

Research Design

A repeated measures experimental design was used to examine the effects of BCAA on lower body power output and perceived muscle soreness following an eccentric exercise bout. All of the subjects visited the lab a total of 5 times over a period of at least 7 days.

CHAPTER II

REVIEW OF LITERATURE

Introduction

On average, the human body is composed of approximately 18% protein². Protein serves as a main component of many tissues in the body including: muscles, organs, glands, enzymes, hair, and other living cells of the body^{2, 17}. It is a complex structure made up of amino acids which can be essential or non-essential. Essential amino acids (phenylalanine, valine, threonine, tryptophan, isoleucine, methionine, histidine, leucine and lysine) must be obtained from the diet while non-essential amino acids can be synthesized in the body ¹⁷. Therefore, humans rely on dietary sources to obtain the essential amino acids necessary to promote growth, repair damaged cells and tissue, and for a variety of metabolic and hormonal activities. A variety of foods contain protein including: lean meats, fish, low fat dairy products, and soy².

Several studies have measured muscle protein balance (synthesis minus breakdown) after resistance exercise^{4, 18-21}. It has been determined that protein synthesis is elevated along with an increase in protein degradation causing a negative balance of muscle protein^{7, 19, 22} until amino acids are provided intravenously or orally^{6, 19, 23}. This stimulation of muscle protein synthesis is reflective of an increased delivery of amino acids to the muscle^{19, 24}. The acute exercise-induced increase in muscle protein synthesis

for up to 48 hours suggests that feeding at any time during these 48 hours should stimulate a greater muscle protein synthetic response compared with feeding at rest¹⁸.

Because of these findings, protein supplementation has been a popular trend in the past years and continues to be one of the most common supplementations used among both recreationally active athletes and competitive athletes^{5, 17}. However, research regarding protein supplementation has been equivocal in almost every aspect including; timing³, amount⁵, and type of protein supplementation used⁵.

Timing of Intake

The timing of nutrient intake is an important principle in optimizing an athlete's performance in order to augment physiological responses to exercise and promote recovery². There is an increase in both muscle protein synthesis as well as muscle protein degradation following a bout of exercise^{7, 22}. In order to stimulate an increase in muscle mass, protein synthesis must be greater than protein degradation²⁵. This also aids in an increased recovery rate¹⁰ which allows for more intense training in a less amount of time.

Pre-Exercise: The results of a study by Tipton et al (2001) indicate that the response of net muscle protein synthesis to consumption of an oral essential amino acid-carbohydrate supplement solution immediately before resistance exercise is greater than when the solution is consumed after exercise, primarily because of an increase in muscle protein synthesis as a result of increased delivery of amino acids to the muscle ²⁶. These results were somewhat contradicted by the same investigators (2007) when the anabolic response to 20 grams of whey protein ingestion was similar whether ingested before or following exercise²⁷. However, no carbohydrates were ingested at the same time as the protein in the previous study. Anderson, et al investigated protein versus carbohydrate

ingestion around an exercise bout. A comparison of the effect of a 14-week resistance training regimen in combination with either carbohydrate or protein supplementation before and after exercise was evaluated. Results indicated resistance training combined with the protein supplementation before and after workouts induced muscle hypertrophy¹⁸. Because of these results and the amount of research supporting post-exercise ingestion of amino acids, discussed below, the ingestion of amino acids is typically focused on following exercise.

Post-Exercise: Tipton et al. (1999) have also investigated the effects of essential amino acid ingestion after an exercise bout. It was concluded that the ingestions of oral essential amino acids results in a change from net muscle protein degradation to net muscle protein synthesis after heavy resistance exercise in humans similar to that seen when the amino acids were infused²⁸. When protein metabolism was studied in human subjects using the stable isotope technique, it was found protein synthesis was increased for 24 hours and even up to 48 hours after one training session in men regularly engaging in resistance training. Protein degradation was also increased and only in combination with nutritional intake was a net gain in protein achieved²⁹. Muscle recovery was further investigated when Etheridge, et al determined that ingesting a single dose of 100 g of protein immediately after an eccentric exercise bout increases the recovery rate of isometric force and dynamic power production during the delayed-onset of muscle soreness in recreationally active males¹¹.

The amount of time allowed before supplementation after a workout has been investigated. When Levenhagen, et al compared supplementation given either immediately after or three hours after an exercise bout, it was determined that the

subjects ingesting the supplement immediately after the exercise bout had a leg protein synthesis increase more than threefold compared to the subjects ingesting the same supplement 3 hours later¹⁸.

Recommended Intake

The current recommended dietary allowance (RDA) for the general population is 0.8 g/kg/day¹. This was initially thought to be adequate for athletes as well¹. The Acceptable Macronutrient Distribution Range for protein intake for adults is 10-35% of total energy¹. The American Dietetic Association has reported that there is not a strong body of evidence documenting that additional protein is needed by healthy adults involved in endurance or resistance exercise, but the Association does state the intake does not recognize the unique needs of competitive athletes¹.

The amount of protein needed for athletes has been of much debate for many years, but research within the last 20 years has indicated that athletes who are involved in intense training need about twice the amount of the RDA in order to maintain protein balance and to increase rate of recovery¹⁴. Over time, a negative protein balance may lead to muscle wasting and training intolerance¹⁴. From this research, it is now generally recommended athletes involved in moderately intense training consume 1.2-1.5 g/kg/day of protein while athletes involved in frequently intense training consume 1.5- 2.0 g/kg/day of protein^{2, 14}. There may be varying protein needs for endurance trained athletes versus resistance trained athletes. The ADA recommends a general standard of 1.2-1.4 g/kg/day for endurance training athletes and 1.6-1.7 g/kg/d for resistance training athletes¹. However, the intensity of the training needs to be assessed as well as special needs of the athlete based on age²¹, sex¹³, and training status¹³. Research has also

suggested that athletes training at high altitude may need even more protein and as much as 2.2 g/kg/day has been found necessary to maintain a positive nitrogen balance ¹.

Types of Protein

Varying amounts of the different amino acids serve as the building blocks of protein. Protein is classified as complete or incomplete protein depending on whether or not the protein contains suuficient amounts of essential amino acids¹⁷. Animal sources of protein contain all essential amino acids and are therefore complete sources while plant proteins are missing some of the essential amino acids and are classified as incomplete protein. Additionally, there are varying levels of quality of protein depending on the amino acid profile of the protein¹⁴. Complete protein sources that contain greater amounts of essential amino acids generally have higher protein quality². High quality proteins are thought to be less susceptible to depletion of essential amino acids and therefore be more available to build and repair tissue, theoretically ².

Protein supplements are a convenient way to obtain high quality protein after workouts and between meals. Some of the most common protein supplements as well as supplements which are investigated most commonly include milk protein, whey protein, casein protein, egg protein, soy protein, and bovine colostrums ¹⁴. As mentioned, essential amino acids must be obtained from the diet while non-essential amino acids can be synthesized in the body. As essential amino acids, the branched-chain amino acids (BCAA) have gained much attention in protein supplementation research^{9, 10, 29}.

The branched-chain amino acids include isoleucine, leucine, and valine. These amino acids account for 35-40% of the dietary essential amino acids in body protein and 14-18% of the total amino acids in muscle proteins¹⁰. The following studies have

demonstrated BCAAs play a very important role in protein metabolism. Garlick and Grant infused one group of rats with all nine essential amino acids and another group with only the BCAA. It was found protein synthesis was stimulated equally by the essential amino acids and the BCAA. This demonstrates that BCAAs are the key amino acids which stimulate protein synthesis⁸. This study was later supported when 3 different groups of rats were infused with either a complete mixture of amino acids, a complete mixture of amino acids minus BCAA, or only the BCAA. The results indicated a mixture of amino acids lacking the BCAA was not effective in stimulating protein synthesis, but the other 2 infusions including BCAA stimulated protein synthesis equally ³⁰. This again supports that the BCAAs are the key amino acids that stimulate protein synthesis³⁰.

Further research has been conducted on the BCAA to determine which of the three is responsible for the increased protein synthesis or if a combination of the three are necessary. Leucine has been found to promote protein synthesis and inhibit protein degradation^{5, 30}. Amino acids are a substrate for muscle protein synthesis. However, in skeletal muscle leucine accounts for approximately 9% of muscle amino acids⁵. These findings suggest leucine is not only a building block of proteins but also a modulator of protein metabolism ². When only leucine was injected into growing rats, no changes in protein synthesis in the gastrocnemius muscle were detected. This indicates that supplementation of leucine alone at a dose of 1 mmol/kg had no detectable effect on muscle protein synthesis³¹.

The branched chain amino acids have also been found to decrease muscle soreness⁹ and increase muscle recovery¹⁰ from eccentric muscle damage. Muscle soreness was decreased after squat exercise with supplementation of only 5 g of BCAA in

both non-active females and males for several days following exercise¹⁰. Jackman et al. demonstrated that muscle soreness was reduced significantly in a period of three days with a supplementation of 7.3 grams of BCAA taken 4 times a day in untrained males⁹.

Etheridge et al investigated 9 recreationally active males and the recovery of delayed-onset muscle soreness (DOMS). Subjects immediately ingested either 100 g of milk containing 40 g essential amino acids or a placebo following a downhill run to induce DOMS. The results indicated a single meal of a protein mixture ingested post-exercise increases the rate of force and power restoration¹¹. Muscle performance recovery from protein supplementation was further supported when a supplement of whey protein hydrolysate was given following an eccentric exercise bout. The consumption of this single dose resulted in a more rapid recovery of muscle force-generating capacity¹². However, neither of the previous studies involved participants who were resistance trained.

Factors Affecting Protein Synthesis

As with any nutrient, there are several factors that can affect protein synthesis. The most common factor researched is the ingestion of protein with carbohydrates. Research has indicated that ingesting a protein and carbohydrate supplement prior to exercise can lessen the degree of muscle catabolism observed during exercise while ingesting protein with carbohydrate following exercise can enhance glycogen and protein synthesis². Theoretically, ingesting carbohydrate and protein following exercise may lead to greater training adaptations¹⁴. Other factors found to affect protein synthesis include the amount of calories consumed, the quantity and quality of the protein ingested, the insulin response to the meal, and the digestibility of the food².

Effect of Training Status on Protein Synthesis

Training status affects the post exercise stimulation of muscle protein synthesis by altering the extent and duration of this process. A trained body will synthesize protein in a shorter time period by returning to basal protein synthesis more rapidly than an untrained body¹³.

It is well known additional protein is needed along with sufficient energy to support muscle growth^{1, 6, 14, 15, 25}. The amount of protein needed to maintain muscle mass may be lower for individuals who resistance-train routinely due to more efficient protein utilization¹⁵. Tang, et al compared mixed muscle protein synthesis between legs in individuals training only one leg. The fractional synthetic rate returned to resting levels quicker in the trained leg than in the untrained leg. This study concluded resistance training weakens the protein synthetic response by shortening the duration for which protein synthesis is elevated³²

Other data suggest that the muscle protein synthesis response to resistance exercise becomes more "refined" with training such that available substrates are directed toward the synthesis of different protein fractions depending on the exercise stimulus^{13, 18}. Hartman, et al investigated the chronic effect of resistance training on muscle protein synthesis in untrained males. Eight, young, healthy males participated in a 12 week full body resistance-training program. Various measures were taken pre-, mid-, and posttesting including nitrogen balance, whole-body protein, urea and creatinine levels. Major findings of this study that 12 weeks of resistance training resulted in a decrease in wholebody protein turnover. However, protein net balance and nitrogen balance were more positive. These results indicated a greater net retention of ingested dietary protein following an intense period of resistance training¹⁸.

Summary

As stated at the beginning, research of protein supplementation has been equivocal in every aspect: timing, amount, type, etc. More research about protein is necessary to continue to close the gap on a better understanding of protein supplementation. Although there is a large amount of literature regarding the BCAAs, they are relatively new to the field of research and lacking much solid support. More studies are necessary to provide consistency among results and to develop a better understanding of BCAA supplementation and their effect on various types of training and recovery. The purpose of the current study is to determine if BCAA ingestion following an acute bout of eccentric exercise can attenuate muscle soreness and improve recovery similarly between resistance trained and untrained men.

CHAPTER III

METHODOLOGY

Participants

Two different groups of males served as the subjects in this single-blind experimental trial. One group consisted of resistance trained males and another group of non-resistance or untrained males. A total of 34 male participants (14 trained, 20 untrained), age 18-30 yrs, were recruited and randomly assigned to one of two subgroups within the trained and untrained groups. Resistance trained males were males engaged in lower-body resistance training at least 2 times a week for at least the previous 8 weeks. Untrained males had not participated in resistance training with the last 6-9 weeks, but may have been recreationally active.

There were two supplementation groups and two placebo groups (one each for untrained, one each for trained). All participants were made aware of the procedures, signed a written informed consent, and completed a general health questionnaire. Participants were able to withdraw from the study at any time. All participants were asked to refrain from any kind of protein supplementation. Subjects were asked to not consume any protein powder supplements, protein bars or any other type of supplementation for at least 6 weeks prior to participating in the study and refrain from lower-body resistance exercise for at least 4 days before testing⁷ as well as throughout the

trial period of approximately 7 days. This study was approved by the Oklahoma State University Institutional Review Board. Approved application number ED10109.

Preliminary Measures/Familiarization

All four groups of subjects visited the lab for preliminary measurements. An informed consent and health history questionnaire were filled out, signed by all subjects and checked by the investigator before subjects were allowed to participate. Subjects produced a baseline measure of power using the TENDO unit according to protocols suggested by Jennings et al. Subjects road a stationary bike at their own speed for 5 minutes each for visit for a warm-up. The TENDO unit cord was attached to the back of a belt placed around the waist of each subject's athletic shorts with the unit placed flat on the floor behind the subject. Power analyses were performed with each subject performing 3 vertical jumps with the highest power measurement recorded. There was one minute of rest between each jump to limit fatigue.

Determination of 1 RM for leg extension and leg flexion exercises were determined following the guidelines of the American College of Sports Medicine for 1-RM testing. The subjects warmed up by completing several sub maximal repetitions. The 1-RM was determined within four trials with rest periods of 3-5 minutes between trials. An initial weight that was within the subject's perceived capacity (50%-70% of capacity) was selected to begin with. Resistance was then progressively increased by 2.5 to 20 kg until the subject could not complete the selected repetition; all repetitions were performed at the same speed of movement and range of motion to instill consistency between trials. The final weight lifted successfully was recorded as the absolute 1-RM ³³. Subjects also rated general muscle soreness on the VAS during this visit for familiarization.

This visit to the lab also served as a familiarization for the eccentric exercise protocol to be done at the next visit. The familiarization involved one lift at less than 50% of their concentric 1-RM in which the weight was lifted by the researchers for the subject to lower. One researcher was located along the edge of the leg extension machine and lifted the weight to 0° full extension. The researcher saying "four" signaled the subjects to begin lowering the weight. The researcher continued counting down to 1 over a period of approximately 4 seconds. The weight was lowered until the subject was at 90° and immediately lifted by the researcher back to the subject's full extension. The subject did not perform any concentric exercise during this protocol. The subjects aimed to control the weight in the downward movement for four seconds⁹. A 3 minute break was allotted between each set⁷. Similar steps took place with the hamstring curl (leg flexion) except the subjects took on the load at 90° and lower to 0° following the commands of the researcher.

Subjects were then randomly assigned to one of the two groups (supplementation or placebo). For the purpose of this paper, the untrained individuals assigned to supplementation will be referred to as UTsupp, the untrained individuals assigned to the placebo will be referred to as UTplac, the trained individuals assigned to supplementation will be referred to as Tsupp and the trained individuals assigned to the placebo will be referred to as Tsupp and the trained individuals assigned to the placebo will be referred to as Tplac. Subjects were provided with the supplement at the familiarization visit to be taken 30 minutes before their next visit.

Experimental Trial

The subjects returned to the laboratory a minimum of two days after the baseline measures and familiarization visit. Subjects consumed supplement drinks (either the

branched chain amino acid supplement or the placebo consisting of flavored water) 30 minutes before exercise, 1.5 hours after exercise, 5.5 after exercise, and 10.5 after exercises⁹. The supplement was distributed to the subjects in small cups with lids and directions for mixing attached along with the time the supplement was to be ingested. The subjects continued to ingest the supplements for the two days following the exercise bout. These supplements were ingested in the morning, between breakfast and lunch, between lunch and dinner, and before bed for a total of four supplements per day⁹. Subjects recorded the time of intake of their supplements on their food logs which were checked by the investigator.

The eccentric exercise bout consisted of 10 sets of 8 eccentric repetitions at 120% concentric 1-RM on leg extension and leg flexion with a 3 minute rest between each set⁷. Power measures and perceived muscle soreness were both measured immediately after eccentric exercise, 24 hours after exercise, 48 hours after exercise, and 72 hours after exercise¹¹.

Visits 2-5 consisted of the subject warming up by completing 5 minutes on a stationary bike and then recording their perceived soreness on the VAS. Subjects then performed their three vertical jumps on the contact mat with the Tendo attached. Before subjects left the lab, they were given their next 4 supplements with the times for them to be ingested written on each cup.

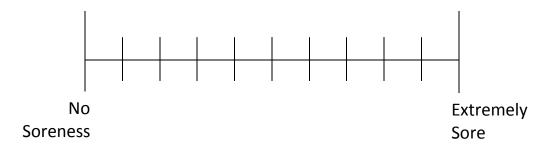
Instruments

The TENDO Weightlifting Analyzer V-207 unit was used to measure power output for vertical jumps being completed. Power measurements were given in watts. The TENDO unit cord was attached to the back waistband of each subject's athletic shorts

with the unit placed flat on the floor behind the subject. This placement allowed for valid readings to be obtained without interfering with jump technique³⁴.

A visual analog scale (VAS) was used to determine perceived muscle soreness by each individual. The VAS consisted of a 100-mm line with anchor points of "no soreness" on the left and "extremely sore" on the right. An example of this VAS is in figure 1. Subjects marked a point along the line to represent the perceived soreness felt in the quadriceps muscle group. Pain was given a value by measuring the distance from the left anchor group to the mark provided by the subjects. It has been determined 90% of pain ratings can be repeated within 9 mm using the VAS⁹.

Figure 1. 100-mm VAS.



Supplement Information

The branched chain amino acid (BCAA) supplements represented a common dosage and nutrient composition that may be found in a typical protein or recovery drink (5-7 g of BCAA/serving). There are no known side effects for consumption of protein at this dosage. The name of the BCAA supplement is Rapid Drive BCAA 5000 and was ordered from the GNC website. This supplement contained the following per serving: 5 calories, 1 carbohydrate, 3000 mg of leucine, 1000 mg of isoleucine, and 1000 mg of valine. The placebo was a generic brand of Crystal Light and contained no macronutrients, but did contain 5 calories as did the supplement. Both supplements were fruit punch flavor. However, there was an obvious difference in texture and taste which was only observed if both the supplement and placebo were tasted. Subjects were only able to see and taste the supplement which they had been randomly assigned so that they were not able to make these comparisons.

Statistical Analysis

Power and muscle soreness were compared among the four groups in order to determine whether BCAA supplementation had an influence on these variables in the days following an intense eccentric exercise bout. A two-way ANOVA (Time x group) was used to identify a significant difference in perceived muscle soreness, peak vertical jump, peak power and average power of the lower body among the four groups (untrained placebo, untrained supplement, trained placebo, and trained supplement). Statistical analyses were performed using SPSS Statistics 17.0 and an alpha level was set of 0.05.

CHAPTER IV

RESULTS

Perceived Muscle Soreness

34 total subjects (14 trained, 20 untrained) completed this study. No subjects withdrew or dropped out of the study once they completed the familiarization/baseline visit. The subject's characteristics are shown in Table 1. A Tukey HSD and Scheffe Analysis of Variance was run among the participant's characteristics to determine if there were any significant differences between the groups. There was a significant difference found between the untrained group receiving the placebo and the trained group receiving the placebo in both their 1-RM for leg extension and 1-RM for leg flexion. However, no differences were found among their age, height, or weight. If weight had been significantly different, this could have had an effect on power outputs.

Table 1. Participants' Characteristics.

	n	Weight (kg)	Height (in)	Age	1RM Leg Ext(lbs)	1RM Leg Flex (lbs)
Untrained Placebo	9	75.6 ± 14.7	69.3 ± 2.9	20.3 ± 2.2	157.8 ± 58.1	89 ± 25.7
Untrained Supplement	11	96 ± 25.7	69.9 ± 4.9	22.6 ± 2.9	170 ± 48.0	104 ± 15.2
Trained Placebo	7	89.9 ± 19.0	69.4 ± 2.4	21.3 ± 1.6	222.3 ± 22.3	119.9 ± 17.1
Trained Supplement	7	81.6 ± 8.3	70.43 ± 1.9	20±1.8	207.1 ± 40.8	99.3 ± 19.7
Total	34	86.4 ± 20.0	69.8 ± 3.3	21.2 ±2.4	185.2 ± 50.9	102.3 ± 21.7

Values are expressed as means \pm SD.

The perceived muscle soreness prior to and following the eccentric exercise bout is illustrated in Figure 1. Visit 1 is the recorded soreness during the familiarization/ baseline visit. Visit 2 was recorded immediately following the eccentric exercise bout, visit 3 is approximately 24 hours after the eccentric exercise bout, visit 4 is approximately 48 hours after the eccentric exercise bout, and visit 5 is approximately 72 hours after the eccentric exercise bout. All groups experienced an increase in perceived muscle soreness following the eccentric exercise bout. However, no statistical differences were found between or within any of the four groups. Soreness ratings tended to be highest 48 hours following the eccentric exercise bout per group.

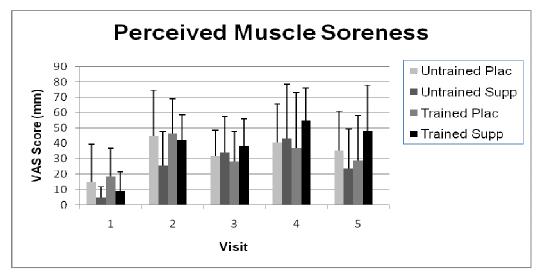


Figure 1 – Perceived muscle soreness before and after an eccentric exercise bout measured using a 100-mm VAS.

Vertical Jump

Of the 3 vertical jumps completed by each subject, the highest vertical jump was entered as the peak vertical jump for that day. There was no significant interaction or main effects of vertical jump for time (p = .406) or group (p = .408). This indicated that there was no increase in jump height for any of the four groups. These results are illustrated in figure 2.

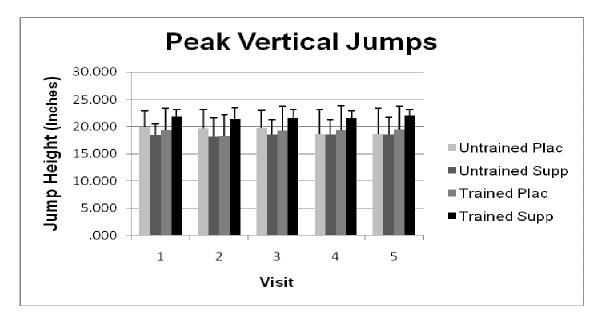


Figure 2 – Peak vertical jumps of the four groups per visit.

Average power

For average power, there was no significant interaction or main effects for time (p = .684); however there was a main effect for group (p = .050). These results are illustrated in figures 3 and 4, respectively. The UTplac had a significantly lower average power than the UTsupp (p = .041). These results indicated that the untrained group receiving the BCAA supplement had a significantly higher average power output than the untrained group receiving the placebo.

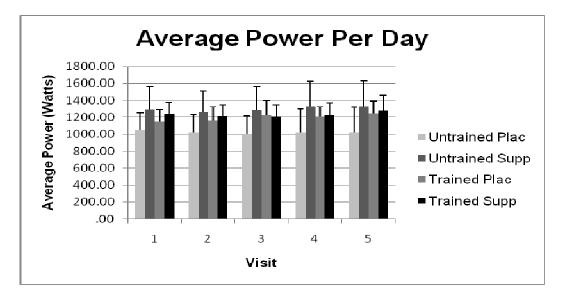


Figure 3 – Lower body average power ouput of the four groups per day .

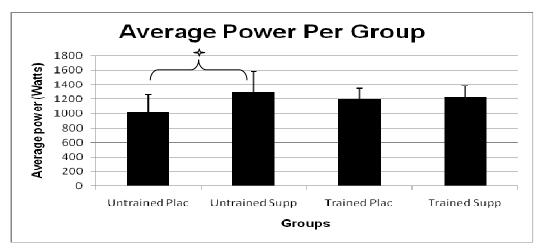


Figure 4 – Lower body average power output per group. \bigstar Indicates significant difference between groups.

Peak Power

For peak power, there was no significant interaction or main effects for time (p = .591) or group (p = .797). These results indicated that there was no difference among the four groups in peak power at any point during the experimental trial. These results are illustrated in figure 5 and indicate there was no difference in peak power regardless of receiving the BCAA supplement.

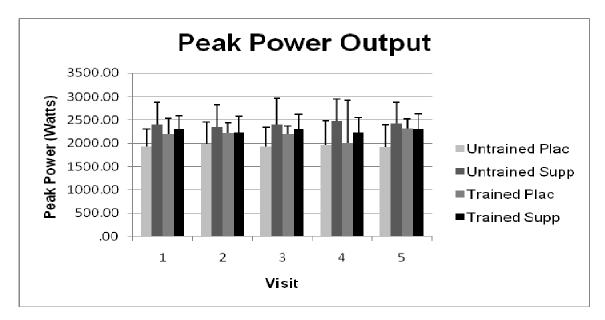


Figure 5 – Lower body peak power output of the four groups per day.

CHAPTER V

Discussion

Power Output

The principal finding of this study was that untrained individuals given a BCAA supplement four times per day for three days following an eccentric exercise bout had a significant increase in average lower body power output compared to untrained individuals receiving a placebo. However, there was no significant difference among the trained participants in average power output in this study. Previous studies have shown an increase in muscle function among recreationally active males following an eccentric exercise bout when given protein supplements^{11, 12}. The present study is the first to compare power and soreness recovery rates following an eccentric exercise bout in trained individuals and untrained individuals.

For this study, there was not a significant difference in average power output between the groups per day. However, the untrained group receiving the supplement had a greater average power and peak power output per day than any other group (figure 3). Comparatively, the untrained group receiving the placebo had a lower average power and peak power output daily than any of the other groups. The trained groups receiving the

supplement had a greater average power output on all days except visit 3 and a greater peak power output on all days except visit 5. Again, there was no significant difference found among the trained groups for peak or average power outputs. Peak vertical jumps were also analyzed for each visit. However, there were no significant differences found. The trained group receiving the supplement did have a higher vertical jump every visit greater than any of the other groups.

Previous studies have found training adaptations to have an effect on protein synthesis⁷. Phillips, et al found the repeated stimulus of resistance exercise appears to reduce muscle protein turnover after exercise. This was demonstrated by an increase in mixed muscle protein fractional breakdown rate in an untrained group with no change in the trained group following a pleiometric exercise bout⁷. Tang, et al compared mixed muscle protein synthesis between legs in individuals training only one leg. The fractional synthetic rate returned to resting levels quicker in the trained leg than in the untrained leg. This study concluded resistance training weakens the protein synthetic response by shortening the duration for which protein synthesis is elevated³². The results of the current study indicate untrained individuals may benefit more from BCAA supplementation than trained individuals in average power output recovery following an eccentric exercise bout due to training adaptations which occur from resistance training.

Perceived Muscle Soreness

Perceived muscle soreness increased in all groups after the eccentric exercise bout. However, no statistical differences were found between or within any of the four groups. Most participants reported the greatest soreness either immediately after the

eccentric exercise or 48 hours post-exercise. Both trained and untrained supplemental groups experienced the greatest perceived soreness 48 hours post-exercise.

The current study used a similar protocol as used by Jackman, et al. However, Jackman, et al found that muscle soreness after eccentric exercise was attenuated in the group receiving the BCAA supplement, suggesting that BCAA are the active ingredient in protein for reducing muscle soreness. A main difference between Jackman, et al and the current study was the amount of BCAA supplementation total. Jackman, et al had subjects ingest 7.3 g of BCAA 4 times a day for three days totaling 87.6 g of BCAA for the trial period. The current study had subjects ingest 5 g of BCAA 4 times a day for three days totaling only 60 g of BCAA for the trial period. This varying amount could be one reason the current study did not have similar results as Jackman, et al.

However, Shimomura, et al also had similar findings as Jackman, et al. Young females and males who did not participate in regular exercise were given either 5 g of BCAA or a placebo and then performed 7 set of 20 squats (amount of weight was not reported). Muscle soreness was evaluated using a VAS before and after the exercise bout as well as the following 4 days. Muscle soreness was highest on the 2nd and 3rd days in the placebo trial. Muscle soreness did increase in BCAA trial, but only peaked on the 2nd day and was significantly lower than that which occurred following the placebo trial¹⁰. This study was different from the current study in several ways: type of exercise performed, amount of reps/sets, amount of BCAA ingested, timing of BCAA ingested, etc. However, the type of exercise used to induce muscle damage and soreness may be the greatest difference leading to the contradicting results. Because Shimomura, et al did not report the amount of weight used to induce soreness, it can be concluded the eccentric

protocol used in the current study induced more muscle damage resulting in more soreness. Therefore, the current study did not see a decrease in soreness as did Shimomura, et al.

It is likely the contradicting results of soreness recovery could be dependent on the subjects from the current study. It is a possibility there may have been some inaccurate reports of soreness by the participants throughout this study. Explanation of soreness was attempted by the investigator. However, many participants expressed great soreness immediately following the eccentric exercise bout. This could have been confused with muscle fatigue. Also, some participants displayed soreness through observed gate by the investigator, but then reported low, if any soreness on the VAS.

Another possibility for not being able to reproduce previous studies results could be because of participants being noncompliant. Participants were informed to not partake in any lower body resistance and more than usual endurance workouts during the experimental trial. However, they were expected to record all physical activity done during the study on their food log. Many participants, with both high and low soreness recordings, did not include physical activity. Therefore, it is not possible to determine, but only assume, workouts they were partaking in affected their perceived muscle soreness.

Conclusions

In conclusion, these findings demonstrate there may be a difference in the way individuals will benefit from BCAA supplementation depending on training status. Nonresistance trained individuals could see a quicker average power recovery rate following

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intense workouts if supplementing with BCAA than those untrained counterparts not supplementing with BCAA.

Limitations of study: The main limitation of the current study was the sample size. It was very difficult to find participants who had been completing resistance exercises at least twice a week for the previous 8 weeks, but were not on protein supplementation. Protein supplementation is a very common trend among the collegiate population which was being recruited from. Other limitations were due to this specific population being non-compliant. Although subjects were asked to not partake in lower-body workouts throughout the experimental trial, many reported doing so on their food records or were seen doing so by the investigator herself. This could have had an obvious effect on the variables being measured and compared. Lastly, the inability to control the subject's dietary intake was a major limitation with this being a supplemental study. Protein, carbohydrates, and total energy intakes could have caused major differences between subjects even within the same groups. Although food records were turned in by most of the subjects, several did not complete them correctly or for all three days they were expected to keep them. Therefore, dietary intake was unable to be assessed properly.

Future Research

Future research needs to be conducted in order to determine if increasing the amount of BCAA supplementation has a greater, positive effect on muscle function recovery. The amount of BCAA could be individualized by assigning a certain amount of BCAA per kg of body weight for the participants as well. Researching a more compliant population may produce different results than the current study found. Lastly, increasing

sample size could produce different values which may lead to significant differences between groups similar to the groups used in the current study.

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APPPENDICES

APPENDIX A

CONSENT FORM

RESEARCH PARTICIPANT CONSENT FORM

"The effects of branched chain amino acids on soreness and power recovery following an eccentric exercise bout in resistance trained and non-resistance trained males."

Principal Investigator

Jennifer Klufa B.S.

Oklahoma State University Department of Health, Leisure, and Human Performance

Purpose of Research

The purpose of this study is to compare the benefits of branched chain amino acid supplementation on the recovery of soreness and power following an eccentric exercise bout in resistance trained and non-resistance trained males.

Procedures

The tasks required of the participant are as follows:

- 1. Complete Health History Questionnaire and Informed Consent forms. The Health History Questionnaire must be completed and if a medical clearance is required, one must be obtained from a physician before participating.
- 2. Attend a familiarization visit where baseline measures will be taken.
- 3. Attend one exercise protocol day followed by one lab visit per day for three, consecutive days in order to collect data.
- 4. Maximum strength (1-repetition maximum or 1-RM) testing will be performed on the lower body using the leg extension and leg flexion exercises with the both legs.
- 5. Subjects will produce a baseline measure of power using the TENDO unit. The TENDO unit cord will be attached to the back waistband of each subject's athletic shorts with the unit placed flat on the floor behind the subject. Power analyses will be performed with each subject performing 3 vertical jumps with the highest power measurement recorded. There will be one minute of rest between each jump to limit fatigue. Power analyses will be completed immediately following the eccentric exercise bout, as well as 24, 48, and 72 hours after the exercise bout.
- 6. Subjects will complete 1 set of three maximal leg extensions and flexions at both 60 degrees per second and 120 degrees per second on an isokinetic machine in order to determine baseline leg extension/flexion torque. These same measures will be taken immediately after the eccentric exercise bout, 24 hours, 48 hours, and 72 hours after the exercise bout.
- 7. Flexibility will be measured and assessed through two tests. The Thomas Test will consist of the subject holding one leg so that the hip is in maximal flexion, while the tested limb will be lowered towards the floor. Measurements will be taken by placing an inclinometer at the midpoint between the patella and the ASIS on the lateral aspect of the quadriceps. Both legs will then be pulled back into the chest and the opposite leg lowered for measurements to be taken. This will be repeated three times with measurements recorded each time.

Okla. State Univ.
IRB
Approved 11/5/10
Approved <u>11/5/10</u> Expires <u>9/14/11</u>
IRB# <u>E.DO9109</u>

The straight leg raise test will also be used to assess flexibility. The subjects will lie on the table with a strap across the waist securing them to the table and with a seat belt fastened across the non-stretched thigh. The inclinometer will then be placed just superior to the malleolus of the ankle and zeroed out. The leg will then be raised keeping the knee straight as the leg is pressed towards the chest. Subjects will notify the investigator when they have reached their stretch threshold. Inclinometer readings will be recorded and then these steps will be repeated three times alternating legs with each reading.

- 8. Two days later, subjects will consume one serving of the supplement (branched chain amino acid supplement or a placebo) randomly assigned to them. Subjects will then partake in a high intensity eccentric resistance training protocol consisting of 5 sets of 8 eccentric repetitions on both the leg extension and leg flexion machine. Eccentric repetitions refer to the lengthening of the muscle and are the opposite of concentric repetitions which are commonly the focus when participating in resistance training. Eccentric repetitions can also be referred to as "negative repetitions". Subjects will consume one serving of the supplement at 1.5 hrs, 5.5 hrs and 10.5 hrs following the exercise protocol.
- 9. The next 2 days will consist of the same schedule. 30 minutes prior to power, torque, flexibility and soreness data being collected, one serving of the supplement will be consumed. Data will be collected at the lab and one serving of the supplement will be taken between breakfast and lunch, another between lunch and dinner, and a final serving before bed. On day 4, subjects will report to the lab for data collection to be taken and no supplement will be taken this day.
- 10. Subjects will be given a Food Log to be kept for four days (the exercise protocol day and the three days following). An explanation of how to use the Food Log will be provided during the familiarization visit.

Visit	Objective	Duration
	1-RM Strength Test: Leg Extension and Leg Flexion	
1	Baseline Measures (Power, Torque, Flexibility)/ Protocol Familiarization	1 hour
	Food Log Explanation	
2	Eccentric Workout Protocol: 5 sets of 8 Repetitions of eccentric work on both the leg extension and leg flexion machines.	1 hour
	Power, Torque, Flexibility and Soreness Measures Assessed	
3-5	Power, Torque, Flexibility and Soreness Measures Assessed	30 min.

Participant visits are described in the following table.

Okia. State Univ. IRB Approved <u>11/5/10</u> Expires <u>9/14/11</u> IRB#<u>E009109</u>

- 11. If pain is experienced at any time, the exercise will be discontinued and the participant will withdraw from the study with no penalty.
- 12. Partcipation in this research is voluntary and there is no penalty for refusal to participate. You are free to withdraw from the study at any time and revoke your consent to participate at any time without penalty.
- 13. A supplementation of Branched-Chain Amino Acids may be ingested throughout this study. The supplement contains a total of 5 g of the branched-chain amino acids: Leucine, Isoleucine, and Valine, per serving. The supplement also contains soybeans. If the Placebo is assigned to be consumed by the subject, flavored water will be ingested containing no macronutrients.

Compensation

No additional incentive is being offered; however individual results following the study may be assessed by the participant upon request.

Risks of participation

Risks associated with the study are minimal; however, due to the nature of the eccentric training protocol muscle soreness may be expected. The muscle soreness poses no more risk than is common from typical resistance training protocols that invoke muscular soreness.

Medical Liability

I understand the risks associated with this study and voluntarily choose to participate. I certify that to the best of my knowledge I am in good physical condition and able to participate in the study. I understand that in case of injury or illness resulting from this study, emergency medical care is available through community health care providers by dialing 911. I understand that no funds have been set aside by Oklahoma State University to compensate me in event of illness or injury.

Confidentiality

Aside from the original data all references will only contain number designations. I will be assigned an ID number. The list of corresponding names and ID numbers will be stored in a locked file cabinet in the Applied Musculoskeletal and Human Physiology Laboratory that only the primary investigator and advisor have access to and will be shredded after data collection has been completed. The health history questionnaire will contain an ID number and will also be shredded following data collection. The written consent will not contain ID number, but will have my signature. This form will be kept in a second locked file cabinet in the Applied Musculoskeletal and Human Physiology Laboratory which only the primary investigator will have access to. It will be shredded three years after the research is completed. Data will be reported as groupings and will not be linked to participants.

Human Subject Statement

If I should have questions about my rights as a research volunteer, I may contact Dr. Shelia Kennison, IRB Chair, 219 Cordell North, Oklahoma State University, Stillwater, OK 74078, 405-744-3377 or irb@okstate.edu.

Okla. State Univ. IRB Approved <u>11/5/10</u> Expires <u>9/14/11</u> IRB#<u>ED09109</u> If I need any additional information concerning the study I may contact Jennifer Klufa 117 CRC, Oklahoma Štate University, Stillwater, OK 74078, 405-744-5507or jennifer.klufa@okstate.edu

Signatures I have read and fully understand the consent form. I sign it freely and voluntary. A copy of this form has been given to me.

Participant's Signature

Date

Investigator's Signature

Date

Ukia. State Univ. IRB Approved 11/15/16 Expires <u>9/14/11</u> IRB# E.D.09/09

APPENDIX B

HEALTH HISTORY QUESTIONNAIRE

ID#:_____

Date:____

Power/Soreness Recovery Supplement Study Pre-Participation Health History Questionnaire

Applied Musculoskeletal & Human Physiology Research Laboratory

Instructions: Write "Y" for yes and "N" for no to all medical problems that you have experienced within one year (unless indicated otherwise).

- _____ History of heart problems, chest pain or stroke?
- _____ Any chronic illness or condition?
- _____ Advice from physician not to exercise?
- _____ Injury to the knee, hip, back, or ankle within the last three months?
- _____ Muscle, joint, or back disorder, or previous injury still affecting you?
- _____ Loss of balance due to dizziness?
- ____ Loss of consciousness?
- _____ Do you smoke or have you quit smoking within the last 3 months?
- _____ Increased blood pressure?
- _____ Difficulty with physical exercise?
- _____ Recent surgery (last 12 months)?
- _____ History of breathing or lung problems?
- ____ Diabetes or thyroid condition?
- _____ Increased blood cholesterol?
- _____ Hernia or any condition that may be aggravated by lifting weights?
- _____ Are you taking any medication for blood pressure or a heart condition?
- _____ Are you allergic to soy products?

_____ History of heart problems in immediate family before the age of 55 years in your father or other male 1st degree relative (i.e. brother, son) or before 65 years of age in mother or other female 1st degree relative (i.e. sister, daughter).

___Other:_____

Checked by (Print):	 Date:	
Signature:		

APPENDIX C

RECRUITING FLYER

Okla. State Univ. IRB Approved <u>11/5/10</u> Expires <u>9/14/11</u> IRB#<u>EDCF1/C9</u>



Power Recovery Study!

Research Participation Opportunity Available

- Participation = 1 week max!
- Possible protein supplementation!
- Become more familiar with the research process!
- Need both resistance trained and non-resistance trained participants!
- Males only
- 18-30 years old
- Must be able to perform leg extension and leg flexion exercises numerous times
- No history of injury (knee, back, hip) within the last 3 months

Study will take place in the Applied Musculoskeletal Human Physiology Research Laboratory in the Colvin Recreation Center.

Contact person: Jennifer Klufa, jennifer.klufa@okstate.edu

Power Recovery Study Jemifer Klufa@okstate.edu Power Recovery Study Jemifer Klufa@okstate.edu	Power Recovery Study Jennifer Klufa jennifer.klufa@okstate.edu Power Recovery Study Jennifer.klufa jennifer.klufa	Power Recovery Study Jennifer Klufa jennifer klufa@okstate.edu Power Recovery Study Jennifer Klufa@okstate.edu	Power Recovery Study Jemnifer Klufa jemnifer.klufa@okstate.edu Power Recovery Study Jemnifer Klufa jennifer.klufa@okstate.edu	Power Recovery Study Jemnifer Klufa jennifer.klufa@okstate.edu Power Recovery Study Jemnifer.klufa jennifer.klufa@okstate.edu
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APPENDIX D

INSTITUTIONAL REVIEW BOARD APPROVAL

Oklahoma State University Institutional Review Board

Date:	Wednesday, September 15, 2010
IRB Application No	ED10109
Proposal Title:	The Effects of Branched Chain Amino Acids on Soreness and Power Recovery Following an Eccentric Exercise Bout in Resistance Trained and Non-Resistance Trained Males
Reviewed and Processed as:	Expedited

Status Recommended by Reviewer(s): Approved Protocol Expires: 9/14/2011

Principal Investigator(s): Jennifer Klufa 117 Colvin Center Stillwater, OK 74078

Douglas Smith 197 Colvin Center Stillwater, OK 74078

The IRB application referenced above has been approved. It is the judgment of the reviewers that the rights and welfare of individuals who may be asked to participate in this study will be respected, and that the research will be conducted in a manner consistent with the IRB requirements as outlined in section 45 CFR 46.

The final versions of any printed recruitment, consent and assent documents bearing the IRB approval stamp are attached to this letter. These are the versions that must be used during the study.

As Principal Investigator, it is your responsibility to do the following:

- 1. Conduct this study exactly as it has been approved. Any modifications to the research protocol
- must be submitted with the appropriate signatures for IRB approval.Submit a request for continuation if the study extends beyond the approval period of one calendar year. This continuation must receive IRB review and approval before the research can continue.
- 3. Report any adverse events to the IRB Chair promptly. Adverse events are those which are
- unanticipated and impact the subjects during the course of this research; and 4. Notify the IRB office in writing when your research project is complete.

Please note that approved protocols are subject to monitoring by the IRB and that the IRB office has the authority to inspect research records associated with this protocol at any time. If you have questions about the IRB procedures or need any assistance from the Board, please contact Beth McTernan in 219 Cordell North (phone: 405-744-5700, beth.mcternan@okstate.edu).

Sincerely.

Philie M. Kennin

Shelia Kennison, Chair Institutional Review Board

VITA

Jennifer Klufa

Candidate for the Degree of

Master of Science

Thesis: THE EFFECTS OF BRANCHED CHAIN AMINO ACIDS ON SORENESS AND POWER RECOVERY FOLLOWING AN ECCENTRIC EXERCISE BOUT IN RESISTANCE TRAINED AND NON-RESISTANCE TRAINED MALES.

Major Field: Health and Human Performance

Biographical:

Education:

Completed the requirements for the Master of Science in Health and Human Performance at Oklahoma State University, Stillwater, Oklahoma in May, 2011.

Completed the requirements for the Bachelor of Science in Nutritional Sciences at Oklahoma State University, Stillwater, Oklahoma in May, 2011.

Experience:

Professional Memberships: International Society of Sports Nutrition (ISSN) and American College of Sports Medicine (ACSM) Name: Jennifer Klufa

Date of Degree: May, 2011

Institution: Oklahoma State University

Location: Stillwater, Oklahoma

Title of Study: THE EFFECTS OF BRANCHED CHAIN AMINO ACIDS ON SORENESS AND POWER RECOVERY FOLLOWING AN ECCENTRIC EXERCISE BOUT IN RESISTANCE TRAINED AND NON-RESISTANCE TRAINED MALES.

Pages in Study: 47

Candidate for the Degree of Master of Science

Major Field: Health and Human Performance

Scope and Method of Study:

Findings and Conclusions: