

The Effect of Two Dosage of BCAA Supplementation on Wrestlers' Muscular Serum Damage Indexes

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Abstract: So far a few studies were done for examining the effect of different dosage of Branched-Chain Amino Acid Supplementation on muscle sermic damage indexes in wrestlers. The purpose of this research was to compare the effects of two dosage of Branched-Chain Amino Acid Supplementation on muscle sermic damage indexes after heavy resistance exercise in wrestlers. 29 young wrestlers were randomly selected and divided into tree groups. All subjects were participated in heavy resistance exercise (3 sets, 10 repetitions, 80% 1RM). The BCAA was given at doses of 210 and 450 mg.kg⁻¹ BW for supplemental groups 1 and 2 respectively, 30 minutes before and after to exercise test and dextrin was given at dose of 210 mg.kg⁻¹ BW for placebo group. To identify enzymes activity (IU/L), venous blood samples were obtained 30 min prior to exercise and at 24 and 48 hrs post exercise. Data were statistically analyzed using ANOVA with repeated measures and Bonfferoni test ($P \leq 0.05$). Based on this study results, CK, LDH, CK_{MB} activity were significantly increased ($p < 0.05$ in all groups. CK, LDH, CK_{MB} indexes having the highest activity in the placebo group, but there were no significant differences between all groups. These results provide evidence that the use of two different dosage of BCAA could not decrease muscle damage associated with heavy resistance exercise.

Key words: BCAA • Muscle damage • Heavy resistance exercise • Wrestlers • CK-CK_{MB}-LDH

INTRODUCTION

Nutritional supplements frequently contain compounds of mainly carbohydrate, protein (essential and non-essential amino acids), vitamins, minerals and another [1-6]. Dietary supplements is very extensive in sport and less athleticism can be found have never experienced in the stages of the championship course to one or more of them [7, 8, 9].

Branched-chain amino acid (BCAA), including leucine, valine and iso leucine that they are classified as essential amino acids. Human body cannot synthesize these amino acids and must be included in the diet [10, 11, 12]. Evidence shows that consumption of branched amino acids has anticatabolic effect during and after exercise [2, 3, 4, 13, 14]. This theory has been proposed that branched amino acid supplements can increase the healing rate is muscle damage after exercise [8, 10, 14-18].

One of the consequences of resistance training is injury, pain and Delayed onset muscle soreness (DOMS). Muscle damage occurs when the muscle cell structure are

breaks [11, 13]. Symptoms of muscle damage is presence of within muscle proteins in the blood, long-term decline in muscle function, including reduction in strength and power, flexibility and muscle dynamic speed [19, 20].

In many researches to measure serum index of muscle damage, such as enzymes, cellular damage and creatine kinase(CK) iso enzymes and lactate dehydrogenize (LDH) has been used [18, 21, 22, 23]. creatine kinase enzymes is considered in phosphate system that is important for energy metabolism in most body cells, especially muscle cells and brain. LDH enzyme found in abundant quantities in the cytoplasm of all tissues or in different concentrations that in the conversion of Pyruvate to lactate or direction anaerobic glycolysis makes up its speed [24-30].

Greer and colleagues (2007) did not see any significant difference between consumption of BCAA and Similar caloric placebo [14]. Zebblin *et al.*, (2007) in double blind study found that consuming eight grams of BCAA before light resistance activity haven't effect on 24 and 48 hours after activity sermic creatine kinase index [31].

Review of conducted researches shows that less research has been done about BCAA supplementation, while the majority of training programs used in this researches, are about the kind of long term and endurance exercises and the results obtained from several studies are not consistent and uniform. So this research question is, taking two different values BCAA supplementation before and after a session of heavy resistance exercises what effect on sermic index of muscle damage (CK-CKMB-LDH) in wrestlers.

MATERIALS AND METHODS

Study Design: The study design was Semi-experimental.

Subjects: Subjects were from trained wrestlers of Mahabad city that volunteers selected according to the criteria and indicators to fitness. Then, according to research objectives and the nature of the research 29 wrestlers (1- low value supplement group, 2- high-value complementary group and 3- placebo group were assigned) were randomly selected.

Sample Selection: For making Homogeneous between groups, the maximum aerobic power, anaerobic power and one repetition maximum (1RM) in the desired movements were measured. Then subjects were classified in three abovementioned groups. Subjects did not practice any sports activity a week before the test and were not used any drugs or supplements also according to terms of medical questionnaires all subjects were healthy. After completing the consent form of design, subjects was forbidden during the research protocol execution taking from any medication, supplements and heavy physical activity. The caliper was used for measuring the skin fat thickness of subjects.

Method of BCAA Supplementation: First BCAA supplement (50 percent leucine, 25 percent iso leucine and 25 percent valine) was prepared to the required amount. Then using Digital Sartovious Scales (models: GM312) with measure accurately 0/1g and 310g capacity, the amount 68 mg/kg for six days before the exercise test and two days after the exercise test, 210 and 450 mg/kg for performance day was placed in a special plastic.

Placebo for this study was Dextrin. Before taking supplements, it use explained to the subjects by researcher, then any of the subjects were required six days, three meals daily (before meals) do consumption

68 mg/kg these supplements. In test day, the supplement group with lower value consumes 210mg/kg, supplement group with a high value consumes 450 mg/kg and placebo group consumes 210mg/kg supplements, 30 minutes before and after the exercise test.

Heavy Resistance Exercise Protocol: To assess the muscular damage, the heavy resistance exercise program was used. At first Multi-joint movements and then single-joint movements were used. Resistance training activities at the 80 percent of 1RM was chosen; in case the subject's have ability to do more than one repetition, the Cochrane formula had been used [4]. Number of movements, including 7 movements with the three sets and ten were repetitions. Rest interval between sets was considered three minutes; rest between movements was one minute interval. Movements including leg presses, chest presses, wire stretch, front thigh, the front arm barbell, bend knees and crunched abdomen.

Blood Sample: Blood sampling was collected in tree time of pre test, 24 h and 48 h after exercise protocol. Measurement method was thus subjects after entering the laboratory, each for five minutes sat on the chair and then by the laboratory technicians, 5 ml blood from anticubital vein were obtained. Subsequently, the blood samples were placed for 30 minutes at a laboratory temperature until clotted. Then separating head by the centrifugal manufactured by Germany Hettich Company and then the amount of enzymes (CK, CKMB and LDH) were measured via auto analyzer device (manufactured by Switzerland COBAS Mira plus company).

Statistical Analysis: Kolmogrov Smirnov test was used for data normality testing. for statistical Analysis, one way analysis of variance with repeated measure ANOVA with between group factor and post hoc Bonferroni tests was used ($05/0p <$).

RESULTS

Subject's physiological profiles to separate the three groups are listed in a table 1.

Research findings show a significant increase in mean and amplitude changes within the group cellular damage serum indexes (CK-LDH-CKMB) within supplementation groups with low value, high value supplements and placebo 24 and 48 hours after the test (Table 2).

Table 1: Psychological profile of subjects in three study groups

Groups variables	Low dosage BCAA supplement group (N=10)	High dosage BCAA supplement group (N=10)	Placebo group (N=9)
Age (years)	1.9±22.40	1.4±22.60	1.6±22.60
Weight (kg)	11.5±73.40	9.2±71.90	12.3±74.40
Height (cm)	4.8±173.8	3.8±174.4	5.3±172.2
Fat percent	2.1±17.20	3.1±17.50	3.7±17.20
Vo ₂ max (ml/kg/min)	4.2±42.40	3.7±43.05	4.01±44.3
Sargent jump (cm)	5.8±52.20	6.2±51.50	7.04±50.8
BMI (kg/m ²)	3.04±24.2	2.93±23.7	4.03±24.5

Table 2: Mean and SD measured (international units per liter) in three sampling period

Activity duration group	Pre activity	24 h after activity	48 h after activity	Change domain after 24 h	Change domain after 48 h
Supplement 1----- CK	29.09±172.4	343.02±78.55	183.5±400.8	324.8±406.01	163.9±228.4
Supplement 2 -----CK	23.9±167.7	214.4±634.1	220.2±468.3	219.4±466.2	220.6±300.6
Placebo -----CK	13.4±177	331.2±762.2	264.04±581.8	322.9±582.2	255.4±404.8
Supplement 1----- CK _{MB}	8.2±19.2	8.5±32.5	6.1±27	8.6±13.3	5.9±7.8
Supplement 2----- CK _{MB}	5.1±2.18	5.7±30	3.5±25	7.6±11.8	5.2±7
Placebo----- CK _{MB}	2.3±19.11	10.1±38.4	8.8±29.4	8.9±19.33	8.2±10.33
Supplement 1----- LDH	29.6±250.3	60.06±394	33.5±311.7	64.3±143.7	32.7±61.4
Supplement 2 ----- LDH	37.9±259.6	72.6±397.9	53.8±354.1	67.7±138.9	62.3±94.5
Placebo ----- LDH	36.9±263.6	61.07±418.6	59.8±344.5	40.2±155	31.1±80.8

Results of analysis of variance with repeated measurements within groups showed that time effect in different time periods (24 and 48 hours after the activity) on (CK-LDH-CKMB) values is significant ($p=0/001$). According to statistical test results, cellular damage indexes in the three groups in LDH (Sig.= 0.734), CK (Sig. = 0.312) and CKMB (Sig. = 0.181) was obtained, which is not significant.

DISCUSSION

Comparison of results between groups in mean and amplitude changes of serum indexes of cell damage (CK-LDH-CKMB), 24 48 hours after the exercise test showed no significant difference between 3 groups. In other words, different amounts of BCAA not significantly affect the serum cell injury indexes (CK-LDH-CKMB), 24 and 48 hours after the heavy resistance activity.

Data Analysis from this study suggest that taking two different values of BCAA cannot have an significant effect on LDH enzyme activity compared with similar calories placebo. After studying Changes in LDH enzyme activity 24 and 48 hours after exercise protocol, a significant increase in activity of LDH was observed in all three groups.

It seems that serum LDH enzyme concentration increases after muscle cell damage in sports activities. When the muscle cell membrane permeability increases or

complete tears occur in muscle cells, enzymes are imported into the blood or lymphatic system [14]. LDH enzyme widely distributed in tissues and its high concentration is found in the liver, myocardial, kidney, skeletal muscle, red blood cells and other tissues. Activity of serum LDH and CK enzymes, like other muscle damage goes up after a period of time, but for long time its concentration remains high [14, 30].

Ferri and colleagues (2006) after running ten sets of ten repetitive plantar flexion motion (to Gastrocnemius and soleus muscles strengthening) with 70 percent of 1RM intensity reported significant increased in LDH enzyme rates [10].

The results of these research is consistent with Greer results (14), whereas is inconsistent with Coombes *et al.*, (2000) and Koba *et al.*, (2007) studies result [7, 18].

For justifying factors affecting the activity of serum enzymes we can refer to fitness levels, muscular type, muscle mass, race and age [4, 5, 17, 23].

Activity of serum enzymes also depends on gender differences. Estrogen hormone has Protective effect on the muscle cell membrane therefore; an increasing amount of serum enzymes in women is less than men. The researches have shown that in resting conditions, CK activity in athletes is higher than non-athletes. So after exercise, less increased seen in athletes serum CK levels [4, 5].

In addition, levels of activity may be increase after taking cholesterol-lowering drugs, asthma, Hypothyroidism, reactive and taking drugs anabolic steroids. Sasaki and colleagues showed resistance exercise significantly increased serum CK for an hour to seven days after the exercise test execution.

The present findings is inconsistent with findings of Coombes and Koba et all studies [7, 18, 19]. Koba and colleagues showed that taking 10 grams of BCAA supplementation compared with placebo reduced serum CK and LDH activities [24]. This inconsistency probably was due to the type of subjects. Also, in this study was used a heavy resistance activity for muscle cell damage. Activities with high and low intensity resistance increased serum CK activity [15]. Reason for the present findings contradict with previous research findings, can be the difference in anabolic hormone response to BCAA intake in endurance and resistance activities.

Results related to CKMB isoenzymes activity levels before and 24 and 48 hours after heavy resistance activity indicates that it is adding a BCAA supplement to diet and two different dosage BCAA taking before and after heavy resistance activity, does not affect activity of CKMB isoenzymes. Increase isoenzymes CKMB 24 and 48 hours after heavy resistance activity in two groups of BCAA supplements was lower than placebo group but, this differences wasn't statistically meaningful.

Range of CKMB isoenzymes changes after 24 h using t-test showed that between BCAA supplementation with high dosage and placebo groups exists difference close to significant level. Likely with increasing the amount of BCAA supplementation or reduction of activity intensity, we could see significant changes.

Vigorous physical activities potentially damaging to cardiac function and aren't essentially harmless. The relative risk for heart cell damage during intense physical activity until about an hour and then increases. Cardiac dysfunction caused by exercise, if there is no cardiovascular disease, shows the category of symptoms that is called heart fatigue.

Atashak (2006) concluded that creatine consumption weekly significantly increased the level of CKMB isoenzymes [1]. Also Faramarzi and colleagues (2007) reported that three sessions of intense periodic Soccer activity is significantly increased CKMB isoenzymes and Carbohydrate supplementation significantly reduced CKMB isoenzymes activity compared with was placebo group [9].

Muscle cell damage has been studied in human and animal models. Most signs of muscle cells damage were

delayed onset muscle soreness (DOMS), which is very dependent on the type and intensity of sporting activity. DOMS is usually emerges eight to 24 hours after cells damage and usually reached to peak 24 to 48 hours after exercise. On the other hand, creatine kinase secretion after 24 hours reaches to peak. Some studies have mentioned that in resistance activities, after 48 hours the amount of creatine kinase secretion reaches maximum.

Many studies in exercise physiology in relation to damage muscle cell, blood sampling is done 4, 8, 24, 48 to 120 hours after activity, we used Zebblin model for blood sampling [31]. The main reason for making the blood, 24 and 48 hours after our activities will returns to accumulation of these enzymes in the blood (after creation of damage). Of course, mechanisms and processes of muscle injury can be effective to justifying our blood sampling method.

Generally, from the results of this study we can infer that the effect of consuming BCAA supplement, particularly with more value, have very poor effect in prevent of increased serum cellular damage enzymes activity. But much research is needed to actually determine the effect of different dosage of BCAA intake on cellular damage serum indexes.

REFERENCES

1. Atashak, S., 2006. The effect of CK supplement on juvenile football players serumic indexes (Persian). University Tabriz, pp: 73-79.
2. Bigard, A.X., P. Lavier and L. Ullmann, 1996. Branched-chain amino acid supplementation during repeated prolonged skiing exercises at altitude. *Int. J. Sport Nutr.*, 6(3): 295-306.
3. Blomstrand, E., P. Hassmen, B. Ekblom and E.A. Newsholme, 1991. Administration of branched-chain amino acids during sustained exercise -Effects on performance and on plasma concentration of some amino acids. *Eur J Appl Physiol Occup Physiol.*, 63(2): 83-8.
4. Brancaccio, P., M. Limongelli and N. Maffulli, 2006. Monitoring of serum enzymes in sport. *Br J Sports Med.*, 40(2): 96-7.
5. Brancaccio, P., N. Maffulli and F.M. Limongelli, 2007. Creatine kinase monitoring in sport medicine. *Br. Med. Bull.*, 81(82): 209-30.
6. Carli, G.M., L. Bonifazi and C. Lodi, 1992. Changes in the exercise-induced hormone response to branched chain amino acid administration. *Eur. J. Appl. Physiol. Occup. Physiol.*, 64(3): 272-7.

7. Coombes, J.S. and L.R. McNaughton, 2000. Effects of branched-chain amino acid supplementation on serum creatine kinase and lactate dehydrogenase after prolonged exercise. *J. Sports Med. Phys. Fitness*, 40(3): 240-6.
8. Cowman, J., 2003. The Effects of Branched-Chain Amino Acid Supplementation on the Exercise Time to Exhaustion in Sedentary Individuals FSU Digital Library Center, 24(2): 102-107.
9. Faramarzi, M., A. Gaeini and R. Kordi, 2007. The effect of periodic burst training and carbohydrate supplement on biochemical heart cells parameters changes of football players (Persian). *Olympic*, 15(3): 21-7.
10. Ferri, A., M. Narici, B. Grassi and M. Pousson, 2006. Neuromuscular recovery after a strength training session in elderly people. *Eur. J. Appl. Physiol.*, 97(3): 272-9.
11. Gaeini, A.A., H. Rajabi, R. Hamedinie and A. Azad, 2003. Scientific and specific criteria of fitness (Persian). *Physical Edu. Staff*, 5(1): 55-67.
12. Ghazizadeh, S., 2005. Surveying the amount of supplement consumption among the body building athletes of Kermanshah province (Persian). Kermanshah University, pp: 82-85.
13. Gleeson, M., 2005. Interrelationship between physical activity and branched-chain amino acid. *J. Nutr.* 135(6): 1591-5.
14. Greer, B.K., J.L. Woodard and J.P. White, 2007. Branched-chain amino acid supplementation and indicators of muscle damage after endurance exercise. *Int J. Sport Nutr. Exerc. Metab.*, 17(6): 595-7.
15. Guzel, N.A., S. Hazar and D. Erbas, 2007. Effects of different resistance exercise protocols on nitric oxide, lipid peroxidation and creatine kinase activity in sedentary males. *J. Sports Sci. Med.*, 6: 417-42.
16. Hayward, R., K.A. Hutcheson and C.M. Schneider, 2003. Influence of acute resistance exercise on cardiac biomarkers in untrained women. *J. Emerg Med.*, 25(4): 351-6.
17. Kashaf, M. and F. Nameni, 2003. The effect of static stretching trainings derived from eccentric contractions on delayed onset muscle soreness (Persian). *Olympic*, 10(3): 12-21.
18. Koba, T., K. Hamada and M. Sakurai, 2007. Branched-chain amino acids supplementation attenuates the accumulation of blood lactate dehydrogenase during distance running. *J. Sports Med. Phys. Fitness*, 47(3): 316-22.
19. Koba, T., H. Koichiro and M. Sakurai, 2005. Effect Of a Branched-chain amino acids supplementation on muscle soreness during intensive training program. *Med. Sci. Sports Exercise*, 37(5): 43.
20. Marginson, V., A.V. Rowlands, N.P. Gleeson and R.G. Eston, 2005. Comparison of the symptoms of exercise-induced muscle damage after an initial and repeated bout of plyometric exercise in men and boys. *J. Appl. Physiol.*, 99(3): 1174-81.
21. Nigam, P.K., 2007. Biochemical markers of myocardial injury. *Indian J. Clin Biochem.*, 22(1): 10-17.
22. Okada, M., M. Bannai, M. Takahashi, *et al.*, 2007. Branched-chain amino acids and alleviation of muscle damage and fatigue in the hyper-running rat Model. *Med. Sci. Sports Exerc. Suppl.*, 39(5): 364.
23. Rahmaninia, F., H. Nikbakht and K. Ebrahim, 2000. The effect of selected training and ibuprofen on delayed onset muscle soreness after burst eccentric contractions (Persian). *Olympic*, 10(1): 33-9.
24. Ratames, N. and R. Hoffman, 2007. Effects of an amino acid/creatine energy supplement on the acute hormonal response to resistance exercise. *Int. J. Sport Nutr. Exerc. Metab.*, 17(6): 608-23.
25. Sasaki, H. and M.E. Matsuo, 2007. The responses of monocyte and neutrophil levels and maximal strength after resistance exercise and training accompanying slight muscle damage. *Med. Sci. Sport Exercise*, 39(5): 122-4.
26. Shimomura, Y., T. Honda and M. Shiraki, 2006. Branched-Chain Amino Acid Catabolism in Exercise and Liver Disease. *J. Nutr.*, 136(1): 250-253.
27. Smith, J., G. Garbutt, P. Lopes and D. Pedoe, 2004. Effects of prolonged strenuous exercise (marathon running) on biochemical and haematological markers used in the investigation of patients in the emergency department. *Br. J. Sports Med.*, 38(3): 292-4.
28. Tang, F.C., 2006. Influence of branched-chain amino acid supplementation on urinary protein metabolite concentrations after swimming. *J. Am. Coll. Nutr.*, 25(3): 188-194.
29. Tom, A. and K.S. Nair, 2006. Assessment of branched-chain amino acid Status and Potential for Biomarkers. *J. Nutr.*, 136(1): 324-30.
30. Vitrako, M., 2007. Biochemical analysis of training programs. 1st ed. Tehran: Samt Publication, pp: 81-9.
31. Zebblin, M., S.M. Sullivan Baier, M. Johannsen and S. King, 2007. Branched-chain amino acid supplementation maintains muscle power following eccentric exercise. *The FASEB J.*, 5(2): 21-32.