Muscle Soreness Responses to Eccentric Exercise:

Effect of Vitamin B6, B9, B12 supplementation

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Abstract. The purpose of this study was to test the effectiveness of vitamin B6, B9, B12 supplement on perceived muscle soreness (MS) to an acute bout of eccentric exercise (EE). Twenty (aged 22.70 ± 1.94 years) healthy volunteers were randomly assigned to either a treatment or placebo groups. All volunteers were prescreened prior to being accepted as a subject. The subjects were instructed to ingest the pills for 4 weeks prior to the EE and for the next four days. All subjects arrived in the morning, resting MS were assessed and then the EE (4 sets of 12 repetitions of elbow flexing with their non-dominant arm). MS were also obtained immediately after the EE, and 2, 6, 24, 48, 72 hrs post-EE. MS increased over time independent of assessment method (palpation, flexion, stationary) and treatment. MS peaked at 48 hrs post exercise independent of treatment in both groups. The data indicated that four weeks of presupplementation and continued treatment after EE with vitamin B6, B9, and B12 had no significant effect on the perceived muscle pain compared to placebo treatment.

Keywords; muscle damage; soreness; pain management; vitamin

1. Introduction

It is well known that exercise of sufficient intensity can induce skeletal muscle damage. Particularly, unaccustomed lengthening type of muscle contractions result in severe muscle damage than concentric exercise [1]. In the days after exercise-induced muscle damage, focal disruption of fiber ultrastructure (e.g., desmin and dystropin) occurs, muscle proteins including creatine kinase are released into the blood, volume

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and circumference of the limb are increased, and delayed onset of muscle soreness (DOMS) appears [2-3].

As muscle soreness is the most debilitating symptom following high-intensity exercise, it may result in impairment in muscle function. And this uncomfortable symptom can negatively affect not only athletes' athletic performance but also the nonathletes' participation in sports [4]. Therefore, maintaining normal muscle function or accelerating recovery from muscle pain is important for both healthy non-athletes and elite athletes.

Ice, ultrasound, nonsteroidal anti-inflammatory medication, massage, and antioxidative vitamin are well known methods to treat muscle damage and DOMS, but it was also reported that these methods have problems with time efficiency, cost, and undesirable side effects [5-10]. On the other hand, vitamin B complexes, as relatively safe supplements to the body, are known to be involved in DNA and protein synthesis associated with recovery form muscle damage [11]. As a water soluble vitamin, usually B vitamins act as coenzymes, that attaches an enzyme and activates or increases its ability to catalyze metabolic reactions.

Among those B vitamins, especially vitamin B6 plays an important role in amino acid and protein metabolism, and it was suggested that this vitamin might be necessary for the production of muscle proteins [11-12]. Vitamin B9, also known as folic acid is necessary for the synthesis, repair, and methylation of DNA and cell division [11]. Recently, it was reported that this vitamin increased the expression of myoblast differentiation, myotube formation, muscular structure related gene expression [13]. Lastly, vitamin B12, also referred to as cobalamin is normally involved in folate metabolism, which in turn relates to DNA synthesis and tissue growth. In addition, it was reported that vitamin B12 has a potential analgesic effect in neuropathic pain [14].

Considering the effects of vitamin B6, 9, and 12 on protein synthesis and analgesic effect, it could be expected that the supplementation of these B vitamins is not only to promote recovery from muscle damage but also to relieve symptoms of DOMS. However, there has been no research to examine the relationship between B vitamin supplementation and muscle damage. With this purpose, current study examined the effect a 4-week vitamin B6, 9, 12 supplementation on the changes of muscle pain induced by eccentric contraction.

2. Methods

A. Subjects

Twenty untrained healthy and college-aged men (aged 22.70 ± 1.94 years, height 173.70 ± 6.12 cm, weight 73.55 ± 10.05 kg) were participated in this study. Every subject was informed of the purpose and process of this study. All participants did not take drugs or vitamin supplements and had no experience in surgery on the elbow joint. This study was approved by the Keimyung University's institutional review board for human subjects.

B. Supplementation

Twenty-nine subjects were randomly assigned to either a placebo group (dextrose mixture with sodium chloride; n=15) or a vitamin B6·B9·B12 supplementation group (n=14). All subjects started to intake placebo or vitamin B6, 9, and 12 from 4-weeks before eccentric exercise and during 3 days after the exercise. The subjects in vitamin supplementation group consumed vitamin B6·B9·B12 (B6: 3mg, B9: 800 μ g, and B12: 4.8 μ g; Garden State Nutritionals, U.S.A) per day. The placebo group consumed two placebo tablets containing 450 mg of glucose and 200 mg of sodium chloride (Fitness farm Inc., Korea) per day.

C. Experimental procedures

Subject's body weight and body composition were measured using InBody 520 (InBody, Korea) before supplementation. Subjects reported back to the laboratory after a 4-weeks supplementation period in the morning after overnight fast and took a rest at least 15 min before obtaining measurements. MS ratings were than obtained, and MS was assessed using a visual linear scale ranging from 1 to 10 in a rested position, through ROM, and in response to palpation. MS scores were obtained at pre-exercise, immediately after, and 6, 24, 48, and 72 hour after eccentric exercise. All subjects performed eccentric exercise (4 sets, 12 repetitions) using their nondominant arm elbow flexors to induce muscle damage at an angular velocity of 20°·s-1 with 60 seconds of rest between sets and 100° of flexion.

D. Statistical analysis

Statistical analysis was used the SPSS statistical data analysis software package (Version 25.0; IMB, U.S.A). Subjects' characteristics was analyzed using an independent t-test to compared between groups. Data for MS was analyzed using 2 x 7 repeated measure ANOVA. Statistical significance was set at $P \le 0.05$, and the data are presented as mean \pm SD.

3. Results

E. Baseline characteristics

All subjects participated in this study were (N = 20) successfully completed all aspects of the testing. There were no statistical differences at baseline between the groups for their characteristics, as listed in Table 1.

	Placebo group (n=10)	Vitamin B group (n=10)		
Age (yrs)	23.10 ± 1.37	22.30 ± 2.41		
Height (cm)	175.00 ± 7.09	172.40 ± 5.02		
Weight (kg)	73.03 ± 8.35	74.06 ± 11.96		
BMI (kg/m2)	23.87 ± 2.68	24.89 ± 3.71		
% body fat (%)	15.15 ± 4.59	19.41 ± 7.94		

 TABLE I.
 PHYSICAL CHARACTERISTICS OF SUBJECTS

F. Muscle soreness results

There were no significant effects on interactions and group main effects in all three different measuring conditions. However, there were significant time main effects, independent of treatment, in MS rating results by three different conditions (at rest, after elbow flexion and extension, and after palpation, $P \le 0.001$). In all three different measuring conditions, MS were significantly increased ($P \le 0.001$) in both group after eccentric exercise, and MS peaked at 48HR after eccentric exercise, independent of treatment.

TABLE II. MUSCLE SORENESS RESPONSES AT REST IN RESPONSE TO ECCENTRIC EXERCISE OVER TIME

	PRE	POST	2HR	6HR	24HR	48HR	72HR	Contrast	
Placebo	0.00	1.17 ± 1.45	1.18 ± 1.60	0.80 ± 1.52	2.15 ± 2.26	2.73 ± 2.36	1.82 ± 1.93	Pre < Post, 2, 24, 48,	
Vitamin	0.00	0.67 ± 0.78	0.66 ± 0.99	0.22 ± 0.34	1.06 ± 1.26	2.44 ± 2.52	1.56 ± 1.95	2, 24, 48, 72HR	
Sig.	Group	Group: F=0.780 (P=0.389), Time: F=11.068 (P=0.000), Group x Time: F=0.437 (P=0.693)							

Note: Values are presented as mean \pm SD. BMI: Body Mass Index. There was no significant difference between the two groups (P ≤ 0.05).

ECCENTRIC EXERCISE OVER TIME									
	PRE	POST	2HR	6HR	24HR	48HR	72HR	Contrast	
Placebo	0.00	3.11	1.88	1.50	4.19	4.81	2.85	Pre <	
		± 1.94	± 1.97	± 1.97	± 2.35	± 2.23	± 1.89	Post, 2, 6,	
Vitamin	0.00	2.09	1.72	1.15	3.36	3.83	3.18	24, 48,	
		± 1.54	± 1.97	± 1.73	± 2.13	± 3.40	± 2.81	72HR	
Sig.	Group: F=0.370 (P=0.550), Time: F=22.200 (P=0.000), Group x Time: F=0.700								
	(P=0.579)								

 TABLE III.
 MUSCLE SORENESS RESPONSES AFTER ELBOW FLEXION AND EXTENSION IN RESPONSE TO ECCENTRIC EXERCISE OVER TIME

TABLE IV. MUSCLE SORENESS RESPONSES AFTER PALPATION IN RESPONSE TO ECCENTRIC EXERCISE OVER TIME

	PRE	POST	2HR	6HR	24HR	48HR	72HR	Contrast	
Placebo	0.00	0.67	0.57	0.24	3.25	3.43	2.54	Dra (Da at	
Flacebo	0.00	± 1.20	± 1.15	± 0.43	± 2.30	± 1.98	± 2.48	Pre < Post, 6, 24, 48,	
Vitamin	0.00	0.64	0.36	0.50	2.33	3.15	2.44	0, 24, 48, 72HR	
		± 1.34	±0.79	± 0.66	± 1.96	± 2.67	± 2.16	/211K	
Sig.	Group: F=0.710 (P=0.685), Time: F=19.238 (P=0.000), Group x Time: F=0.344								
	(P=0.792)								

4. Conclusion

The present study examined the effect of a 4-week vitamin B6, B9, B12 supplementation on muscle soreness (muscle pain) induced by acute eccentric exercise. As a main finding of this study, vitamin B complex treatment compared with a placebo treatment for 4 weeks before and continued for 4 days after the eccentric exercise did not attenuate the increases in muscle soreness although MS scores in the vitamin B group tended to be low at the all measurement time points after eccentric exercise. These results suggest that 4 weeks of pre-supplementation and continued treatment with vitamin B6, 9, 12 at the level of twice the recommended nutrient intake had no significant impact on improving muscle pain associated with muscle damaging procedure.

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