Available online at www.scholarsresearchlibrary.com

Scholars Research Library

Annals of Biological Research, 2013, 4 (8):163-168
(http://scholarsresearchlibrary.com/archive.html)

ISSN 0976-1233
CODEN (USA): ABRNBW

Effects of zinc, magnesium and vitamin B_6 supplementation on hormones and performance in weightlifters

Nader Moëzzi, Maghsoud Peeri and Hasan Matin Homaei

Department of Exercise Physiology, Central Tehran Branch, Islamic Azad University, Tehran, Iran

---

ABSTRACT

The purpose of this study was to determine the effect of one period of Zinc, Magnesium and vitamin B_6 supplementation on testosterone and cortisol concentrations and ratio of testosterone to cortisol and performance in weight lifters. To this end, 12 weightlifters were selected among weightlifters of Kamyaran city and after completing questionnaire, were randomly divided into two groups: ZMA group (n=6; age: 18.76±2.39 year; height: 171.00±7.64 cm; weight: 74.36±10.01 kg) and placebo group (using dextrose) (n=6; age: 20.17±2.13 year; height: 171.66±3.77 cm; weight: 78.39±9.37 kg). Both groups were trained for 7 weeks (preparatory season training for weightlifters) 6 days per week and all had the same condition. Double-blind procedure was performed. Fasting blood samples 6 cc of the antecubital vein in the sitting position of subjects before and after completion of the course and in the same were collected. To measure total testosterone, free testosterone and cortisol, ELISA method was used. Vertical jump and long jump tests, medicine ball push test, squats test and military presses test were used to assess subject's performance. Data' analysis was done using student t-test particularly dependent and independent groups; also, significance level was set to be $p \leq 0.05$. Results: no significant differences in the rate of total testosterone ($p=0.695$), free testosterone ($p=0.379$), cortisol ($p=0.346$) and ratio of testosterone to cortisol ($p=0.718$) and strength in both groups were improved. Conclusion: it seems that athletes to obtain more advantages in this field of sport do not need to use this supplements.

Key words: ZMA supplement, hormones, performance, weightlifters.

INTRODUCTION

It has been shown that exercise performance capability in intensive physical activities is affected by nutrition quality and quantity, vice versa [1]. Nowadays, dietary supplementation along with exercises is performed and a few athletes can be found that in their sporting life does not use one of them [2]. There are different types of supplements on the market and it is claimed that their use is beneficial for athletes, but there are only a limited number of them which their usefulness in improvement of athlete's performance has been proved scientifically [3]. Thus, supplementation may help to rectify negative effects of dietary deficiencies and improved athlete's performance [3]. Such estimated that at present, the cost of dietary supplements is more than 20 billion dollars. Cassileth (2009) reported that in 2009, 52% of people use 1 or more than 1 type of supplements, which in comparison with consumption level of about 46% in 2006, indicating an increase in supplement usage[4]. There is a study that mentioned advantages of dietary supplementation [5].

ZMA is a special mixture of zinc, magnesium and vitamin B_6. In this product zinc and magnesium are exist as zinc aspartate and zinc mono-L-methionine, and magnesium aspartate, respectively. Actually, zinc and magnesium are combined with aspartate in order to increase of their sorption [6].

Zinc is an intracellular cation, which has catalytic and structural functions [7]. This element is involved in the structure of many metabolic enzymes which are needed for natural digestion and maintenance of normal hormonal levels [8]. Zinc deficiency has unfavorable effects on testosterone (anabolic hormone), and consequently on muscular mass and strength [8]. Decrease of zinc may intensify during exercise (long-term activity with very high sweating) and due to inadequate consumption [9]. It was reported that zinc is existed in the structure of carbonic anhydrase and lactate dehydrogenase, which are involved in metabolism during athletic exercise [10] and zinc deficiency is high in athletes and persons, they have recreational activity [1, 7]. It has been mentioned that zinc decrease can result in damaging to immune system and decrease in performance [3, 11].

However magnesium is an element that has a very determining role in many cellular reactions, it has involved in more than 300 enzymatic reactions that in them, food is catalyzed and new chemical products are formed [12], such as: glycogen breakdown, fat oxidation, protein synthesis, ATP synthesis and secondary messengers system. Indeed, magnesium is performed as a physiological regulator of membrane permanency and has important roles in immune, hormonal and heart performance [7]. It is clear that magnesium is decreased levels of cortisol hormone [8]. Excessive levels of cortisol have a catabolic effect on skeletal muscles [8]. Therefore magnesium supplementation can lead to control cortisol levels, which increases muscular mass and strength [8]. It has been shown that vitamin B_6 has a series of compounds that are found in muscles, which are uniformly involved in releasing of glucose from glycogen [13]. These compounds have important roles as cofactor in many cellular reactions related to energy metabolism. Severe shortage of these compounds can lead to impaired energy production during hemoglobin synthesis. Use of it can stimulate production of growth hormone [13].

Fatigue during exercise (especially intense exercise) is due to several factors, one of which is production and accumulation of ammonia in the body. Excess ammonia, which is a by-product of exercise, is excreted as urea by aspartates [14]. The combinations of minerals with aspartic amino acid are called ‘mineral aspartate’. Aspartic acid function is a transitional mechanism for minerals that increase their sorption [8].

Results of conducted studies by Brilla & Conte (2000) on the effects of ZMA supplementation on serum levels of anabolic hormones and strength of football players showed a 30% increase of total testosterone (Tt) and free testosterone (Ft) hormones and a 4% increase of IGF-1 hormone and significant increases in power and strength in supplement group, while in placebo group there were a 10% decrease in serum levels of Tt and Ft, and a 22% decrease of IGF-1 hormone. Also, improved sleep quality and significant increase in strength and power was reported in supplement group [11].

In the study of Wilborn *et al*., (2004), the effects of ZMA supplementation on training adaptations, and anabolic and catabolic symptoms were examined. They were found no significant difference between treatment and placebo groups and there were no improvement of training adaptations in individuals with strength training. These researchers by examining levels of Tt, Ft, IGF-1, cortisole, growth hormone, ratio of testosterone to cortisol (T/C), and improvement of training adaptations has found no significant difference between two groups [10]. In another study, the levels of serum testosterone and urinary excretion of steroid hormone metabolites after administration of a high-dose zinc supplement were studied by Kohler *et al*., (2007), and the results showed that supplementation was increased significantly zinc level of serum, urinary excretion of it, and urine volume, while there was no change in serum Tt and Ft levels in response to that supplementation [3].

According to factors such as the use of dietary supplements among athletes of different sports and specially ZMA supplementation among athletes of strength sports in the hope of finding desirable replacements for anabolic steroids, as well as, conflicting results and comments of conducted researches on the ZMA supplementation, and lack of specific investigation of ZMA supplementation effects on weightlifters, the present study was done. So the purpose of this study was to determine the effects of 7 weeks ZMA supplementation on levels of testosterone and cortisol, ratio of testosterone to cortisol (T/C), and performance (leg and shoulder muscles power and strength) in weightlifters.

MATERIALS AND METHODS

- Subjects and study plan

Among weightlifters of Kamyaran city, 12 people who qualify to participate in this study (including not having any symptoms of illness, injury, lack of drugs and supplements during at least in 3 past months) were selected as subjects. At first, the objectives of the study were explained to them in detail. Then, in order to obtaining personal characteristics including weight, height and fat percentage of the subjects were measured. After completing the questionnaire, subjects were randomly divided into two groups of placebo (dextrose) and ZMA supplement, each with 6 persons. Both groups were trained for 7 weeks (preparatory season training for weightlifters) 6 days per week and all had the same condition, except that the placebo group used dextrose. Fasting blood samples prior to manipulate independent variables (training program and ZMA supplement) and after completion of the course in the same situation were collected. Muscular power and strength tests (in order to determine one repetition maximum) were performed by subjects before and after conducting this training design. Vertical jump (Sargent) and long jump tests (leg muscles power), medicine ball push test (shoulder muscles power), squats test (Quadriceps femoris muscle strength) and military presses test (shoulder muscles strength) were used to assess subject's performance. Pre- and post-test amounts of these parameters were measured.

- Blood samples

Fasting blood samples of 6 cc were collected from antecubital vein in the sitting position of subjects before and after completion of the study course at the same condition (at 8 AM following 12 hours fasting and 48 hours after last training session). Collected blood samples were kept in test tubes for 60 min and after clotting of them, they were centrifuged in 4000 rpm speed for 15 min. Then the serum samples were collected and stored at -20°C until hormone analysis.

- Hormone analysis

Measurements of Tt and Ft were performed using Monobind kit made in USA with a sensitivity of 0.038 ng ml^{-1}, and IBL kit made in Germany with a sensitivity of 0.1 pg ml^{-1}, respectively. Also, levels of cortisol were measured using IBL kit made in Germany with a sensitivity of 2.46 ng ml^{-1}. ELISA method and STATFAX2100 apparatus were used for evaluation Tt, Ft and cortisol levels.

- Training program

Training program were done in the preparatory season for weightlifters. This program was ranged from 3 to 6 movements per training session, 2 to 6 sets for each movement and 1 to 6 repetitions per set. The duration of rest periods between sets and movements were varied respectively between 2-4 and 3-5 min. Training program was performed 7 weeks, 6 sessions per week and exercise intensity was fluctuated between 50 to 130% according to the type of movements and exercise volume (table 1).

Table 1. one of the training programs of weightlifters

Movement	Number of sets	Number of repetitions	Intensity
Hang power clean	1-3	4	80% 1RM power clean
Hang power snatch	3	4	70% 1RM power snatch
High pull	3	2	110% 1RM clean and jerk
Back squat	3	3	80% 1RM back squat

- Supplements

In this study, the ZMA supplement made by Ultimate Nutrition Company of USA was used. This product was prepared as capsules in boxes, each contains 90 capsules. All of the subjects of supplement group used 3 ZMA capsule per night after eating dinner and before bedtime, so that all of three capsules totally contains 30 mg zinc mono methionine aspartate, 450mg magnesium aspartate, and 10.5 mg vitamin B_6. The subjects of placebo group were used a substance (dextrose) that was seemingly similar to ZMA supplement, but was ineffective. Nutrition of subjects was controlled with regard to energetic needs of them by planning a dietary program. The diet contains 15% protein, 60% carbohydrate and 25% fat [14].

- Statistical analysis

First Kolmogorov-Smirnov test was used to ansure normal distribution of data. Paired simple t-test was used in order to comparison of averages of each parameter before and after treatments. Significant differences between supplement and placebo groups were assessed using independent t-test. The significant level was p≤0.05 for all the calculation and all the statistical tests were conducted using SPSS software(version 13, Michigan, USA).

RESULTS

Subject's characteristics are presented in table 2.

Table 2. Measured parameters (mean ± SD) for supplement and placebo groups include age, height, weight, fat percentage and BMI.

Variable	ZMA supplement Group	Placebo Group
Age (year)	18.76 ± 2.39	20.17 ± 2.13
Height (cm)	171 ± 7.64	171.66 ± 3.77
Weight (kg)	74.36 ± 10.01	78.39 ± 9.37
Fat percentage (%)	16.91 ± 5.59	19.14 ± 3.27
BMI (kg m^{-3})	25.33 ± 1.54	26.62 ± 3.33

Measured levels (mean ± SD**)** of Tt, Ft, cortisol and (T/C) ratios in different treatments are showed in table 3. There were increases in levels of Tt and Ft at the end of the study period than their initial levels in both groups, but there was no significant change within groups. A significant decrease (P = 0.027) was occurred in cortisol level of supplement group. While there were significant increase (P = 0.355) and decrease (P = 0.486) in T/C ratio respectively in supplement and placebo groups, but the increase in supplement group at the end of study period was not significant. Following 7 weeks supplementation, no significant decrease in levels of Ft (P = 0.379), Tt (P = 0.695), cortisol (P = 0.346) as well as T/C ratio (P = 0.718) between supplement and placebo groups were observed.
Mean measured Values of Sargent and long jump tests (leg muscles power), as well as medicine ball push test (shoulder muscles power) were increased after study period in both groups, but levels of these parameters showed no significant change at intergroup level (table 4). There was significant increase in mean measured value of squats test (Quadriceps femoris muscle strength) after completion of the training course (p=0.025 for ZMA group; p=0.004 for dextrose group). Also, significant increases (p=0.011 for ZMA group; p=0.010 for dextrose group) were observed in mean measured value of military presses test (shoulder muscle strength) after study period. Comparison of measured values of each parameter between ZMA and dextrose groups showed no significant difference between these groups.

Table 3. Measured variables (mean ± SD) and P values for levels of free testosterone, total testosterone, cortisole and testosterone to cortisole (T/C) ratios

Group	variables	Pre-test	Post-test	P value
Exercise and ZMA supplement	Free testosterone (pg ml^{-1})	8.45 ± 3.18	9.54 ± 3.94	0.683
	Total testosterone (ng ml^{-1})	4.38 ± 1.43	5.75 ± 2.11	0.432
	Cortisole (ng ml^{-1})	156.33 ± 23.82	129.50 ± 22.18	0.027*
	Testosterone to cortisole ratio	51.89 ± 15.16	68.01 ± 27.92	0.355
Exercise and placebo	Free testosterone (pg ml^{-1})	5.48 ± 1.85	5.86 ± 2.26	0.681
	Total testosterone (ng ml^{-1})	3.95 ± 1.34	5.01 ± 1.78	0.181
	Cortisole (ng ml^{-1})	113.66 ± 42.84	106.30 ± 41.96	0.559
	Testosterone to cortisole ratio	46.71 ± 5.99	57.64 ± 14.87	0.486

**significant difference between pre- and post-test values in supplement group.*

Table 4. Pre- and post-test measured amounts (mean ± SD) of performance tests vertical jump (Sargent) and long jump tests (leg muscles power), medicine ball push test (shoulder muscles power), military presses test (shoulder muscles strength) and squats test (leg muscles strength) and P-values for supplement and placebo groups.

Group	variables	Pre-test	Post-test	P value
Exercise and ZMA supplement	Vertical jump (Sargent) test	57.00 ± 11.66	59.16 ± 12.44	0.273
	Long jump test	239.33 ± 35.89	240.50 ± 37.16	0.482
	Medicine ball push test	495.33 ± 58.65	499.00 ± 55.31	0.546
	Military presses test	62.58 ± 16.41	67.71 ± 19.00	0.011*
	Squats test	155.86 ± 48.63	163.50 ± 45.47	0.025*
Exercise and placebo	Vertical jump (Sargent) test	50.16 ± 9.53	51.33 ± 8.47	0.499
	Long jump test	215.66 ± 36.91	219.00 ± 32.81	0.231
	Medicine ball push test	500.16 ± 55.35	503.50 ± 49.17	0.685
	Military presses test	59.98 ± 14.93	64.65 ± 15.68	0.010*
	Squats test	130.30 ± 28.18	134.95 ± 26.72	0.004*

**significant difference between pre- and post-test values in supplement group.*

DISCUSSION

The results of prior studies on ZMA supplementation effects on testosterone and cortisole levels are not aligned and in some cases are at odds to each other [3, 10, 11]. Brilla and Conte (2000) studied the effects of ZMA supplementation on serumic levels of anabolic hormones and strength of football players and they reported significant positive effects of this supplement(11). Their findings showed 30 and 4% increases in serum levels of (free and total) testosterone and IGF-1 respectively in supplement group, while there were 10 and 22% decrease in levels of testosterone (free and total) and IGF-1respectively in placebo group. In study of Wilborn *et al*., (2004), the effects of ZMA supplementation on training adaptations and anabolic and catabolic profiles were investigated. The results of their study showed no difference in serum levels of free and total testosterone, IGF-1, cortisol, growth hormone and T/C ratio between experimental and placebo groups. Also, there was no significant effect during exercises related to training adaptations in subjects with strength training [10]. Kohler *et al*., (2007) reported that use of ZMA supplement significantly increases zinc content of serum and urinary excretion of it, as well as the urine volume, but is not lead to change total and free testosterone in response to supplementation [3].

The results of present study showed that 7 weeks ZMA supplementation was lead to 11.42 and 21.36% increase respectively in free and total testosterone than initial levels, while the levels of these hormones in placebo group has shown 6.48 and 21.15% increases, respectively. However, our results showed increase of testosterone levels within each group, there were no significant differences between initial and final levels of testosterone hormone within groups, and between two groups at the end of the study period. The results of Wilborn *et al*., (2004) and Kohler *et al*., (2007) studies are supported our results. In the other hand, results of Brilla and Conte (2000) are in contrast to the our results, because they reported 30% increase in total and free testosterone in supplement group, as well as 10% decrease in these hormone's levels in placebo group. The lack of significant differences may be a result of regular training of participants and since the same increases were occurred in free and total testosterone levels in both groups, so we can not attribute that increases to ZMA supplementation.

Other results of this study showed that, cortisol levels in supplement and placebo groups were decreased respectively 17.16 and 6.47% at the end of study period than initial levels. Decrease of cortisol concentration was significant only in supplement group ($P = 0.027$). The results of the study of Wilborn *et al*., (2004) also showed no significant difference in cortisol level due to 8 weeks ZMA supplementation along with 4 session of strength training per week between supplement and control groups, which supports our results[10].

Our findings indicated that use of ZMA supplement along with preparatory season exercises for weightlifters was lead to significant decrease of cortisol level in supplement group, while there was no change in cortisol level of placebo group. It has been shown that magnesium decreases cortisol levels [8]. Considering that magnesium is one of the compartments of ZMA supplement, the significant decrease in cortisol concentration can be attributed to ZMA supplementation. Cortisol concentration is one of the factors influencing on testosterone levels. In fact, T/C ratio represents anabolic-catabolic activity of the body [15]. T/C ratio must be considered in investigation of testosterone level and it's adaptation during resistance training [16]. Testosterone to cortisol (T/C) ratio has been suggested as a measure of Anabolic to catabolic (An/Cat) status during resistance training. Therefore, increase in testosterone level and decrease in cortisol concentration or both of them probably indicates tissue anabolism [17].

Power index was assessed by investigating the power of shoulder muscles (using medicine ball push test) and leg muscles (using Sargent and long jump tests). Our results showed that the power of shoulder muscles in medicine ball push test were increased 0.74% and 0.66% in ZMA and dextrose groups, respectively. Also, the power of leg muscles in Sargent test were improved 3.78% in ZMA group and 2.33% in dextrose group, as well as the leg muscles power in long jump test were improved 0.48% and 1.54% respectively in ZMA and dextrose groups. These increases in power of shoulder and leg muscles were not significant and in the all of tests, there were no significant difference in power of subjects between two groups. Brilla and Conte (2000) reported that eight weeks supplementation were lead to significant difference between supplement and dextrose groups that is inconsistent with our results. In the other hand, the results of the study of Wilborn et al., (2004) showed no significant difference in power between supplement and dextrose groups after eight weeks ZMA supplementation along with resistance training (4 sessions per week), which is supported our results. These insignificant differences between intergroup data can be related to the effects of regular training of subjects and is not the effect of ZMA supplementation.

Muscular strength variable was studied by investigating the strength of shoulder muscles (military presses) and leg muscles (squats). Our results showed that shoulder muscles strength in military presses in ZMA and dextrose groups were increased significantly ($p<0.011$) 8.3 and 7.78%, respectively. Intergroup differences in military presses were significant in both groups; therefore the differences between pre- and post-training records cannot be due to supplementation and it may be the result of regular training course.

Leg muscles strength (squats) were significantly ($P <0.025$) increased 4.9 and 3.56% in ZMA and dextrose groups, respectively, but there was no significant difference in squats records before and after training course in ZMA and dextrose groups. Although, differences in ZMA and dextrose groups were significant, these differences can be attributed to the effects of regular and continuous training.

In this study, no significant difference was observed in strength of shoulder muscles (military presses) and leg muscles (squats) between ZMA and dextrose groups after training period. The results of Wilborn *et al*., (2004) and Koehler et al., (2007) supported the results of our study, but Brilla and Conte (2000) reported the results that are incompatible with our results.

CONCLUSION

In the present study the effects of ZMA supplementation on hormones and performance (leg and shoulder muscles strength, leg and shoulder muscles power) in weightlifters was investigated and based on the obtained results despite of the significant differences in Testosterone, Cortisol ,T/C, ,power and strength between supplement and placebo groups, ZMA supplementation was lead to improvement of anabolic hormones levels, decrease in cortisol concentrations, increase in T/C ratio and improve power and strength levels of subjects that can be useful. Our results showed that weightlifters don’t need ZMA supplementation for obtaining further advantages in specific trainings of weightlifting.

Acknowledgement
The authors would like to acknowledge all of the weightlifters of Kamyaran city for their presence in this study.

REFERENCES

[1] Nielsen, F.H., Lukaski, H.C. *Magnesium Research*, **2006**, 19 (3), 180-189.
[2] Amirsasan, R., Nikookheslat, S., Sari-Sarraf, V., Kaveh, B. *World applied sciences journal*, **2011**,13(6), 1544-1548.
[3] Koehler, K., Parr, M.K., Geyer, H., Master, J., Schanzer, W. *European Journal of Clinical Nutrition*, **2007,**63, 65-70
[4] Driskell, J.A., Wolinsky, I. Nutritional Assessment of Athletes, 2nd ed., CRC Press. Taylor and Francis Group, **2011**; p.393.
[5] Dib, P. M.Sc. thesis, Université de Balamand, 2006.
[6] Adele, S., Jalali, R. Sports supplement buyers guide., Basic Health publication, **2007**; pp.94,141,236.
[7] Lukaski, H. C. Am. J. Clin. *Nutri*, **2000**, 72, 585-593.
[8] Antonio, J. and Stout, J. R. Supplements for strength-power athletes., Human Kinetics Publishers, **2002**; pp.123,125,149.
[9] Cordova, A., Navas, F.J. Ann. *Nutr. Metab*, **1998**, 41,274-282.
[10] Wilborn, C.D., Kerkisick, C.M., Campbell, B.I.m Taylor, L.W., Marcello, B.M., Rasmussen, C.J., Greenwood, M.C., Almada, A., Kreider, R.B. *Journal of the International Society of Sports Nutrition*, **2004**, 1(2), 12-20
[11] Brilla, L. R., Conte, V. *Journal of exercise physiology*, **2000**, 3(4), 26- 36.
[12] Shils, M.E., Magnesium. In: Shils, M.E., Olson, J.A., Shike, M., eds. Modern nutrition in health and disease. 8th ed. Philadelphia: Lea & Febiger, **1993**; pp.164-184.
[13] Catherine G. Ratzin Jackson. Nutrition and the strength athlete., CRC Press, **2001;** pp.86,103.
[14] Kleiner, S. M. Power eating, 2nd ed., Human Kinetics publishers, pp. 144,217
[15] Hakkinen, k., pukarinen, A., Alen, M., komi, p. *Eur.J.Appl.physiology*,**1985**,53, 287-293.
[16] Kramer, W.J. *Medicine and scince in sport and exercise*, **1988**, 20, 152-157.
[17] Komi, P.V. Strength and power in sport., Volume III. 2nd ed. Blackwell Science Ltd, **2003**; P. 523.