

## **The Effect of 6 Weeks of High-Intensity Exercise Training on Serum Calcium of Martial Arts Women**

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### **Abstract**

**Purpose:** Cell homeostasis is an essential factor in maintaining cellular health and intracellular mechanisms, which can also be associated with inflammatory factors and increased stress for muscle cell health. Calcium is also a necessary factor in cell health and is to establish homeostasis in the body. This study aimed to investigate the effect of high-intensity training on serum calcium levels in combat women. **Method:** For this purpose, 18 women karateka in the age range of 17 to 23 years were randomly divided into 2 groups: control (n=7) and exercise (n=11). The exercise group performed proper exercises to improve karate and aerobic techniques for 6 weeks in the intensities of more than 80% of the maximum heart rate reserve. Physiological indices and blood samples were measured 24 hours before and 48 hours after the training protocol. To measure calcium cell biomarker, the Pars Azmoon calcium kit was used. Research data were analysed by covariance test at the significant level ( $P \leq 0.05$ ) using SPSS software version 25. **Results:** After six weeks of high-intensity training, there was a significant difference in serum calcium levels in the exercise group compared to the control group ( $P = 0.002$ ). Also, the results within the group showed a significant increase in serum calcium ( $P = 0.016$ ) in the exercise group compared to baseline conditions. **Conclusions:** High intensity training seems to increase serum calcium in female athletes, and this can lead to precursors to cellular disorders, muscle damage and increase inflammatory factors and ultimately decrease the performance of female athletes. However, more research is essential in this regard. **Keywords:** Calcium, Martial Arts, Muscle Injury, Intensity Exercises

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## INTRODUCTION

Calcium (Ca) is the most abundant mineral in the human body. It has also been shown that exercise can make a significant difference in blood calcium levels (Ramazanpour et al. 2010). Calcium also plays a significant role in cell health, including the nerves function, blood coagulates, cardiac function, and muscle contractions. There are three distinct fractions of calcium in the blood: free calcium (48%), protein-bound calcium (40%), and calcium bound to inorganic anions (12%). Increased levels of calcium ions in the blood cause factors such as weakness, lack of energy, anorexia, nausea and vomiting, constipation, abdominal pain, and urination in large quantities. Calcium release during stress and executes by Translokon (proteins involved in translation). In martial arts, muscle injury is known. Because karate is a collision sport, it exposes athletes to more injuries by using foot and hand kicks (Shirvani et al. 2015). Karateka women with properties less muscle mass, higher fat percentage, and hormonal changes, are more vulnerable than Karateka men. Hence, one of the serious discussions in the science of karate practice is the culmination of necessary world-class sports skills. Usually, karate women try to reach the peak of performance before the main competitions by doing high-intensity exercises. With insufficient alternation between performances and rehearsals and with short stages of reconstruction, they are subjected to over-training. Therefore, this type of training intensity and stress in the peak of performance, has been accepted by trainers, leading to increased markers and indicators of muscle damage and physiological stressors in karate athlete. (Azizi et al. 2011). A study has shown that a period of karate competitions significantly increases muscle damage indicators of LDH and CPK levels. (Namani et al., 2004). These biomarkers affect increasing calcium before and even after strenuous activity in health and continued exercise and competition. In intense karate training, due to the depletion of intracellular adenosine triphosphate, changes occur in the function of the calcium pump, possibly disrupting the transport of this ion and increasing intracellular calcium ions. As intracellular calcium concentrations increase, calcium-dependent proteases (calmodulin and calpain) are activated, leading to increased signaling of inflammatory factors (Single Lamperchet 2004).

As, Jing et al. (2011) reported that 8 weeks of resistance exercise by mice increased calcium enzyme concentrations. Also, some researchers reported that six weeks of resistance exercise insignificant increased calcium levels Parhampour et al (2013) . In this relation, higher concentrations of total calcium and calcium ionization in active people have been reported compared to the control group (Maimoun L., and Sultan C., 2009).

However, lampchert et al. (2004) reported a decrease in intracellular ATP and dysfunction of the calcium pump, and therefore an increase of intracellular calcium concentration. Given the biological importance of calcium in establishing intracellular homeostasis and muscle contractions, as well as stimulating some of the cellular signaling pathways, the question arises as to how intense karate training increase the serum calcium concentration in women who practice karate. Therefore, the aim of the present study was to evaluate the serum calcium concentration during 6 weeks of intense training in karate women.

## **METHOD**

### **Subjects**

The statistical population of the present study consists of practicing karate girls in the age group of 17 to 21. Subjects randomly divided into two groups of exercise (n = 11) and control (n = 7). The subjects filled out health and physical activity questionnaires. They were examined by the team doctor, and after obtaining the inclusion criteria, including having one of the bronze, silver, or gold medals in international or national competitions, no history of drug use, regular participation in karate training, no history of long-term injury and no use of analgesics and inflammation drugs that affect serum calcium concentrations, participated in this study. Exclusion criteria is included sports injuries such as ruptured ligaments and meniscus, fractures, dislocations during exercise and additional exercise outside the training protocol, leaving the camp, irregular participation in exercise, and taking anti-inflammatory drugs and calcium supplements or Supplements affecting the concentration of calcium ions are mentioned. Eligible subjects were given full explanations (written and oral) about the purpose of the research, manner of conducting the exercise training, conditions for

participating in the plan, blood sampling, practical methods, benefits, and possible disadvantages. Also, the method of completing the nutrition questionnaire and the nutrition self-report form were presented. Finally, the subjects completed and signed a written consent form before starting the exercises and were said that they could leave the research program at any time without any obligation. This research was approved by the ethics committee of Allameh Tabataba'i University with the code IR.ATU.REC.1399.013.

### **Physiological and anthropometric characteristics**

Physiological and anthropometric variables, such as height (cm) and weight (kg) were measured with a height gauge and a digital scale made by Novin brand made in Iran and with an accuracy of 0.1., as well as body mass index (kg / m<sup>2</sup>), fat content (percentage) with Inbody 370s, resting and training heart rate (beat/minute) with polar clock, blood pressure (mm Hg) with German Madison MTS model and maximum oxygen consumption (ml/kg/min) was estimated by Bruce test.

### **Exercise protocol**

The training protocol consisted of 12 training sessions of 45 to 90 minutes per week for 6 weeks. Each session consisted of warming up, performing training patterns, and cooling down. karateka women performed the newest exercise training of TT , TD , Randori, HIIT and technical training (Figure 1) (J. Ravir 2009) (Tabben M 2014) . The exercise group performed the Karate exercises in the morning and the afternoon, with an intensity of more than 80% of the maximum reserve heart rate. Also, the recovery period was about 3 hours after each training session. The control group followed their daily life and did not participate in the training protocol. But they had the same conditions of nutrition, rest, and sleep.

exercises protocol	TT training	technical training
		2 times per a week
	TD training	technical development training
		2 times per a week
	randori trainin	an individual ,fre 15min warm au period.seven repitations of 2min fre karate sparring bout separated by 2min passive recovery were performed.
	2 times per a week	
aerobic and agility training	fied exercises and body weight training	
	3times per a week	
strenght training	free weight and machine esercises	
	3times per a week	

**Figure 1.** Karate exercises protocol in elite women karateka

### **Dietary and medication intake measures**

Information was collected from each individual based on a structured and previously validated ( $r = 0.89$ ) self-administered questionnaire. The self-administered questionnaire ascertained sociodemographic characteristics, medical history, and lifestyle habits, such as caffeine consumption. The questionnaire also included a 100-item modified block food frequency questionnaire (FFQ) (Huang et al., 2002) . This FFQ estimates average daily nutrient intake, including total calories, protein, fat, carbohydrate, vitamins, antioxidants, and

micronutrients, based on questions about frequency and portion size of a given food and usual eating habits over the past year. The FFQ took about 30 min to complete. Further, standard and self-reported questionnaires were used to collect information on the use of medications and supplements during the period of study (Maleki and Tartibian, 2018).

### **Blood sampling and biochemistry**

The blood sampling stage was performed under the supervision of trained personnel. 8 cc of fasting venous peripheral blood samples at 08:00-09:00 were collected in fasting conditions 24 hours before the start of training and 48 hours after the end of the six weeks exercises. To separate serum, blood samples after clotting for 15 minutes at room temperature, were centrifuged at 3000 rpm for 15 minutes at 4 ° C and serum calcium levels (mg/dl) were assessed by the standard automatic autoanalyzer -BT 1500 system by Pars Azmoun Biochemistry Kit.

### **Statistical analysis**

Results are expressed as mean  $\pm$  standard deviation (SD). Shapiro1 test was used to determine the natural distribution of data. ANCOVA analysis of covariance was used to test the research hypotheses. SPSS software version 25 was used for data processing at the significance level ( $P < 0.05$ ).

## **RESULTS**

### **Baseline characteristics**

In the Ex group, body fat percentage, systolic blood pressure, and resting heart rate decreased ( $p < 0.05$ ) but, maximum heart rate and  $Vo_{2max}$  significantly increased ( $p < 0.05$ ). Also weight and diastolic blood pressure none significantly decreased after 6 wks of high-intensity exercises in the Ex group.

**Table 1:** Baseline characteristics of Karateka women in pre and post test of six wks high intensity exercise.

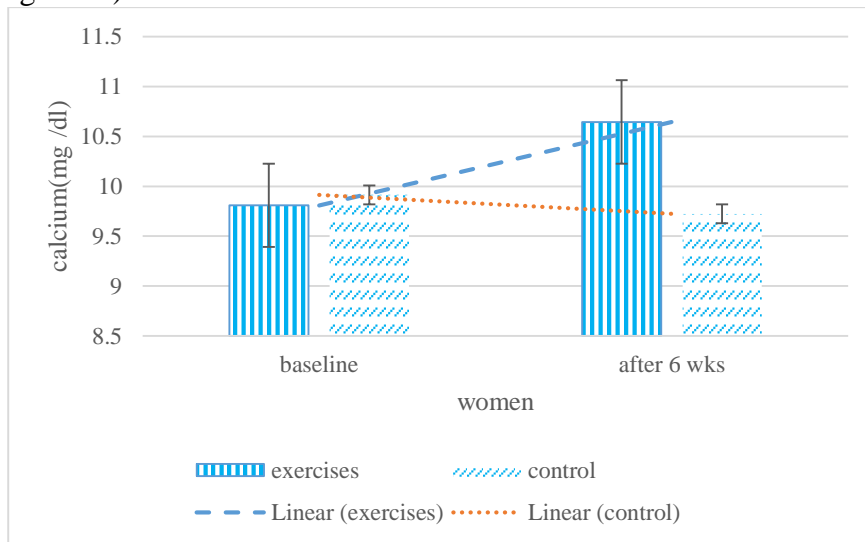
		Pre exercise	Post exercise
Variable	Group	Mean $\pm$ SD	Mean $\pm$ SD
Age (year)	EX	18/63 $\pm$ 1/2	18/63 $\pm$ 1/2
	C	18/85 $\pm$ 1/34	18/85 $\pm$ 1/34
Weight(kg)	EX	52/27 $\pm$ 7/73	52/36 $\pm$ 6/94
	C	55/00 $\pm$ 4/89	55/14 $\pm$ 4/91
Height (cm)	EX	162/36 $\pm$ 5/14	162/72 $\pm$ 5/001
	C	161/71 $\pm$ 4/6	161/85 $\pm$ 4/67
SBP(mmg)	EX	110/63 $\pm$ 2/24	*108/54 $\pm$ 2/2
	C	110/71 $\pm$ 2/57	111/28 $\pm$ 2/75
DBP(mmg)	EX	76/09 $\pm$ 1/13	75/72 $\pm$ 1/27
	C	75/85 $\pm$ 1/34	76/14 $\pm$ 1/21
Fat %	EX	18/90 $\pm$ 2/21	*18/27 $\pm$ 2/49
	C	18/42 $\pm$ 2/71	18/85 $\pm$ 1/86
RHR(beat per min)	EX	60/31 $\pm$ 2/52	*58/18 $\pm$ 2/45
	C	60/42 $\pm$ 2/63	61/42 $\pm$ 2/43
MHR(beat per min)	EX	200/00 $\pm$ 1/67	201/00 $\pm$ 1/18
	C	200/42 $\pm$ 1/71	199/28 $\pm$ 1/38
VO2 max (ml kg per minute)	EX	42/18 $\pm$ 1/60	*43/61 $\pm$ 1/43
	C	42/42 $\pm$ 1/9	41/57 $\pm$ 2/22

\*: p<0.05, Mean  $\pm$  SD: Mean and standard deviation

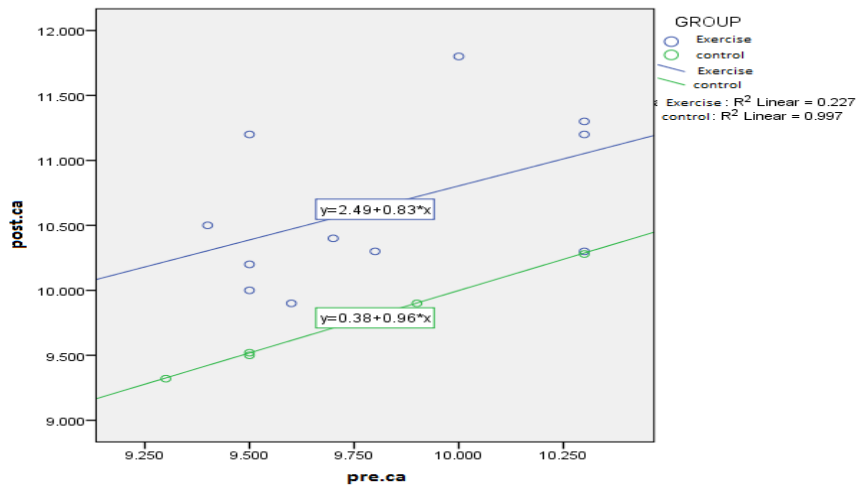
### Serum calcium

After six weeks of karate training of competition season, there was a significant difference in serum calcium levels ( = 0.496 and P = 0.002) in the exercise group compared to the control group. (Figure 2) Also, the within-group results showed a significant increase in serum

calcium levels ( $P = 0.016$ ) only in the exercise group compared to baseline conditions (Figure 2). In addition, there was a linear relationship between the auxiliary random variable and the serum calcium levels (Figures 3).



**Figure 2.** Serum Calcium levels of trained karateka girls, following six weeks of high-intensity exercises



**Figure 3.** Dispersion of the linear relationship between the auxiliary random variable and the calcium dependent variable (mg/dl)



## DISCUSSION

The results showed that 6 weeks of high-intensity karate exercises increased serum calcium of karateka women, and this change correlated with an auxiliary random variable. This data clearly showed that the high intensity of Karate exercises leads to cellular stress. The results of this study also showed that physiological and anthropometric factors changed after 6 weeks. Fat percentage, blood pressure, and resting heart rate decreased after 6 weeks of high-intensity exercise of karate, and maximal heart rate and maximal oxygen consumption increased. researchers reported the increase of serum calcium and decrease of physiological characteristics (Fat percentage, blood pressure, and resting heart rate) after 6-12 weeks high-intensity exercises (Watani et al. 2018, Abdullah Al-Ansreh et al. 2018, Mokhtari and Daryanush 2015, Mohammad Hassani et al. 2015, Ramirez Wells et al. 2020, Parhampour et al. 2013; Jing et al. 2011; and lampchert et al. 2004). To date, no reports have conducted to examine the calcium response to high-intensity karate exercise training. However, there is some evidence that high-intensity karate exercise training induces the creation of cell stress and inflammatory biomarkers (Parhampour et al. 2013). It seems in the effect of high-intensity karate exercises, the excessive reduction of adenosine triphosphate can increase calcium. It disrupts the function of the calcium pump and increases the calcium ion inside the cell. During high-intensity karate exercises, these disorders disrupt the cell's metabolic system and homeostasis, leading to cell destruction and apoptosis. As this condition persists, stress on the endoplasmic reticulum is activated in muscle cells, and inflammatory factors are activated along the way Jakku et al. 2018 . Muscle damage, as the most common injuries, occur in high-intensity martial arts and causes a significant increase in LDH, CPR , and CRP levels (Namani et al. 20045, , and Kadwaki H. 2013) . It is reported that High-intensity training can directly destroy muscle cells through increasing calcium (Kadwaki 2013). But Mal'andish et al. 2016 , has reported no change in serum calcium There is a supposed that Caspases are involved in changes in calcium due to physical activity through the ER pathway. In the ER pathway, caspase-3 is activated through an ER-located caspase, caspase-12 (Nakagawa et al., 2000) . Caspase-12 is predominately situated at the cytosolic side of the ER membrane, and is specifically activated by disturbance to

ER homeostasis, such as ER Ca<sup>2+</sup> depletion. The activation of caspase-12 has been shown to depend on a Ca<sup>2+</sup>-dependent protease calpain (Nakagawa and Yuan, 2000). In addition, the ER can regulate the apoptotic signaling through IP<sub>3</sub> receptor (IP<sub>3</sub>R) or ryanodine receptor (RyR) mediated by Ca release <sup>2+</sup> There is this guess that with increasing serum calcium the health of athletes is endangered, and the role of inflammatory and stressors in the continuation of the path should be examined by exercise physiologists.

## CONCLUSIONS

In conclusion, the results of the present study show serum calcium is affected by high-intensity karate exercises, and it increases significantly. Little researches have been studies on response such exercises on karateka women. These observations suggest that the high-intensity karate exercises could be taken into consideration when investigating cell stress responses to exercise training in Karateka women. It remains to be determined how changes in calcium levels of karateka women function may be connected with cell stress outcomes in these athletes.

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