Original Research Article

Effect of One Session of Resistance Training with and without Blood Flow Restriction on Serum Levels of Creatine Kinase and Lactate Dehydrogenase in Female Athletes

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ABSTRACT
Introduction: Intense exercise may cause immunological changes and muscle damage. The aim of this study was to investigate the effect of one session of resistance training with and without blood flow restriction on serum levels of creatine kinase (CK) and lactate dehydrogenase (LDH) in female athletes. Materials and Methods: In this semiexperimental study, 30 female basketball players were randomly divided into a traditional resistance training group (without blood flow restriction), a resistance training group (with blood flow restriction) and a control group. Blood samples were taken before the first session and 30 minutes after the last training session. T-test and one-way analysis of variance were used for evaluation of within group and between group differences, respectively. All statistical analyses were performed in SPSS (version 23) at significance level of 0.05. Results: Serum level of CK increased significantly in all group, while serum LDH increased significantly only in the experimental groups when compared with pre-test levels (P<0.05). There was no significant difference in serum levels of CK and LDH between the experimental groups and the control group. Conclusions: Resistance training with blood flow restriction and high-intensity resistance training without blood flow restriction have similar effects on markers of muscle damage.

KEYWORDS: Resistance Training, Blood Flow restriction, CK, LDH

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INTRODUCTION
One of the most important goals of physical education and sports science research is identification of suitable exercises for fulfillment of physical fitness plans and athletic skills. One of the issues that arise in this area is exercise intensity that occasionally leads to injury among athletes and detach them from the sport environment. Coaches and athletes have always been seeking new ways to improve and optimize performance. It is well known that high-intensity exercise poses an acid-base challenge to the body. Additionally, it has been reported that pre-exercise acidosis affects anaerobic metabolism and power output during a 4 km time trial (1). Training can also influence the ability to handle the acid-base challenges faced during high-intensity exercise (2). Hence, implementing the most suitable trainings with the least possible damage is one of the most important principles in sport.

Resistance training has gained popularity over the last decade or so as researchers discovered the many benefits it has to offer. It is known as the most effective training intervention for increasing muscle strength (3) and is often recommended for prevention of injury, musculoskeletal rehabilitation, reducing risk of falling and increasing functional capacity (4,5). In order to achieve these goals, intensity of 70% and 85% of maximum repetition has been recommended by the American College of Sports Medicine (6). However, studies have shown that the metabolic-mechanical stress caused by high-intensity exercise training results in undesirable changes in indirect indicators of cellular damage and increases the plasma concentration of metabolic-mechanical stress indices, such as creatine kinase (CK) and lactate dehydrogenase (LDH) (7). In normal state, CK and LDH release into the blood is inhibited by the cell membrane. However,
rupture of the cell membrane, induction of enzyme synthesis and cell proliferation, and increased cellular degradation increase the rate of their release into the bloodstream (8). Several factors including age, gender, type, physical fitness and intensity of exercise affect the level of LDH after exercise. The glycolytic enzyme LDH is a biological marker for cytosol in various tissues (9), and plays a role in glycolysis pathway, production of ATP and cellular metabolism. The enzyme is secreted due to structural changes in the muscle tissue after vigorous activity (10). CK is an enzyme of the phosphagen system, which is important for energy metabolism in most cells of the body, especially muscle and brain cells. CK is a reliable indicator of muscle membrane permeability, since the enzyme is found only in the skeletal and cardiac muscles. Increased concentration of CK in blood may be a sign of muscle damage and inflammation (11). Therefore, it is necessary to design safe and effective exercise trainings for the elderly, patients and other individuals who require muscular strength but cannot tolerate high-intensity exercises.

Some studies have shown that a low-intensity training program with vascular decrease pressure on the joints and ligaments and causes less tissue damage while stimulating muscle strength (12,13). Such trainings are known as Kaatsu training, in which the arterial blood flow and the active muscle blood flow are reduced (12). Even in short term and at low intensities, these trainings can increase muscle strength and cause significant muscle hypertrophy (14). In addition to improving muscle strength and muscle hypertrophy, resistance training increases growth hormone secretion, lipolysis and bone formation, consequently reducing obesity and risk of diseases of the skeletal system. In Kaatsu training, the blood flow in active muscles is limited or blocked during exercise by strapping a rubber flexible cuff to the proximal part of the arm or thigh (15). The intensity of these trainings typically ranges between 20% and 30% of one-repetition maximum (1RM), which is roughly equivalent to the intensity of daily activities. Therefore, people with different physical characteristics can tolerate this type of training (16, 17). Studies have shown that these types of exercises are associated with hypoxia, and reintroduction of blood flow may lead to the production of reactive oxygen species and significant tissue damage (18). In this regard, Abdelfattah et al. studied the effects of 12 weeks of exercise training with limited blood flow in 20 swimmers and found no change in the LDH levels (19). According to medical statistics, females are at higher risk of developing aging-associated diseases compared to males. Therefore, resistance training should be part of females’ exercise program. However, limited information is available on the safety of these trainings on cardiovascular function, muscular trauma and oxidative stress. Thus, the aim of the present study was to investigate the effect of one session of resistance training with and without blood flow restriction on serum levels of CK and LDH in female athletes.

**MATERIALS AND METHODS**

This was a semi-experimental study with a control group and a pretest-posttest design, which received approval from the ethics committee of Shahid Chamran University (code: IR.SCU.REC.1396.24.3.77896) and the Iranian Registry of Clinical Trials (code: IRCT20171203037718N1). Study population included female basketball players aged 23-30 years with 7-10 years of experience who live in the Khuzestan Province, Iran. A physician examined all subjects in terms of drug use, general health, cardiovascular health and blood pressure prior to entering the study. Then, the subjects were randomly divided into three groups: traditional resistance training without blood flow restriction (TRT, 80% 1RM), resistance training with blood flow restriction (RTVO, 30% 1RM) and control group (with blood flow restriction without any exercise training). Three days before the first training session, written consent was taken from the subjects. During
training sessions, correct movements, proper breathing and especial strength was calculated using the Brzycki formula:

\[
IRM = \frac{\text{rep wt}}{[102.78 - 2.78 \times \text{reps}]} 
\]

In the same session, height, weight and body mass index were measured while subjects were fasting and on empty bladder. In order to measure amount of LDH and CK, 5 ml venous blood samples were taken from the subjects’ right hand in the sitting position. The samples were centrifuged for 10 minutes at 3500 rpm to separate serum, which was later stored at -70 °C. Level of LDH was measured by the photometric method using Pars Azmun kits (Iran) with a coefficient of variation of 3.2%. CK level was measured by the enzymatic colorimetric method using Pars Azmun kits (Iran) with a coefficient of variation of 1.2%. Sampling was done before the first training session and 30 minutes after the last training session. The two exercise groups performed one session of resistance training (barbell curl) at the body building club of Shahid Chamran University of Ahvaz, Iran. The training session started with a 5-minute warm up. Armband was strapped on the upper arm of subjects in the

considerations were properly practiced by the subjects and the maximal muscle groups with blood flow restriction. Each armband included a bag that was attached to a manual barometer. The exercise (barbell curl) started with a set of 30 repetitions and ended with two sets at fatigue with a 30 seconds rest time. In all three sets, the pressure of the cuff was set at 120 mmHg. At rest intervals, cuff pressure was relieved. Descriptive statistics were used to calculate central and dispersion indicators. Normality of data was confirmed with the Shapiro-Wilk test. Paired t-test was used to examine within-group differences. The effectiveness of the protocol was assessed using one-way ANOVA. All statistical analyses were performed in SPSS (version 23) at significance level of 0.05.

RESULTS
Some anthropometric characteristics and physical composition of the subjects are shown in table 1. Based on the results of the Levene’s test, there was no significant difference in the measured factors between the three groups.

| Variable                  | Control group | TRT 80% 1RM  | RTVO 30% 1RM | P-value |
|---------------------------|---------------|-------------|-------------|---------|
| Age (years)               | 26.40±0.70    | 25.40±0.54  | 26.18±0.50  | P=539   |
| Height (Cm)               | 169.20±0.98   | 169.60±1.78 | 170.40±1.70 | P=857   |
| Weight (Kg)               | 67.70±1.11    | 60.66±1.87  | 66.70±1.27  | P=4553  |
| Body mass index (Kg/m2)   | 23.65±0.14    | 23.05±0.12  | 23.19±0.23  | P=547   |
| VO₂max (ml/Kg/min)        | 42.90±1.40    | 41.50±0.92  | 40.09±0.79  | P=326   |

We compared pretest and posttest serum levels of CK and LDH in the experimental groups and control group. As shown in table 2, serum level of CK increased significantly in all groups, while serum LDH level increased significantly only in

The experimental groups (P<0.05). Based on the results of one-way ANOVA, there was no significant difference in the CK and LDH serum levels between the experimental groups and the control group.
Table 2. Comparison of pretest and posttest levels of serum CK and LDH between the experimental groups and the control group

| Index     | Groups   | Pretest      | Posttest     | Within Group P-value | Between Group P-value |
|-----------|----------|--------------|--------------|-----------------------|-----------------------|
| CK (ng/ml)| TRT      | 123.30 ±1.76 | 132.25 ±8.27 | 0.022                 | 0.550                 |
|           | RTVO     | 128.20 ±6.99 | 143.60 ±8.15 | 0.012                 |                       |
|           | Control  | 136.60 ±5.92 | 141.90 ±6.34 | 0.048                 |                       |
| LDH (IU/L)| TRT      | 327.50 ±5.06 | 347.30 ±8.58 | 0.001                 | 0.225                 |
|           | RTVO     | 329.10 ±12.28| 351.50 ±10.54| 0.001                 |                       |
|           | Control  | 362.70 ±10.88| 366.20 ±9.18 | 0.434                 |                       |

DISCUSSION

Extreme physical activity exerts biological damage to the phospholipid membrane by inducing a mechanical-metabolic stress. In addition to high mechanical stress, exercise-induced hypoxia may also damage membrane of muscle cells. In our study, there was no significant difference in the serum level of CK and LDH between the two experimental groups and the control group. However, serum levels of CK and LDH increased significantly in all groups compared to the baseline. The results of this study are consistent with the results of studies by Foroughi Parandjuny et al. (20) and Pettersson et al. (21). Foroughi Parandjuny et al. reported that one session of resistance training at 75% 1RM increased muscle damage and perceived muscular pain in male student athletes (20). Pettersson et al. claimed that CK, LDH, aspartate aminotransferase, alanine aminotransferase and myoglobin increased significantly and remained unchanged up to seven days after resistance training (weightlifting) in 15 healthy subjects without resistance training experience (21). Although several hypotheses have been proposed for the mechanism of exercise-induced muscle damage and muscle soreness, the role of lactic acid, muscle spasm,

Connective tissue damage, inflammation and swelling are more profound. It is believed that eccentric contractions in all individuals, irrespective of age, gender or level of fitness can cause muscle cramps but have a greater effect on those who have not performed enough resistance training (22). In addition, muscle-tendon damage following intense eccentric exercise leads to accumulation of calcium and disruption of ATP production. In the next few moments and in the inflammation stage, the number of circulating neutrophils increases with damage. Within 6 to 12 hours of damage, macrophages accumulate in the affected area and produce active histamine. The accumulation of histamine, potassium and quinine due to phagocytosis and cell necrosis raises local temperature and triggers pain receptors within the muscles and tendons. All of these can lead to the release of CK, LDH, aspartate aminotransferase and resulting in feeling of distress and muscle soreness. Increasing body movement in this state can elevate the pain (22).

The results of this study are inconsistent with the results of Takarada et al. (18), Fujita et al. (7) and Hosseinikak et al. (23). Takarada et al. stated that one session of strength training in male athletes does not significantly change CK concentrations (18). The lack of a
significant change in the metabolic-mechanical stress indices are sometimes attributed to the previous experience of the subjects in strength training and exercise. Hosseinikak et al. found no significant change in the indices of muscle damage after one session of training with and without vascular occlusion in young inactive females (23). This result could be due to the effect of 17 β-estradiol as well as a reduction in muscle membrane damage caused by this hormone. Fujita et al. also found no significant change in the metabolic-mechanical pressure indices (7). It has been argued that a short-term exercise program with blood flow restriction is responsible for the lack of response to metabolic-mechanical stress during exercise because serious and often irreversible damage to the skeletal muscles usually occurs after prolonged exposure to ischemia. The lack of a significant change in muscle damage markers can also be attributed to the time of sampling after the last exercise session. Some researchers believe that muscle cramp caused by muscle damage, reaches its peak 24-72 hours after training (24). Thus, it is recommended to collect blood samples for longer periods after the last training session. It should be noted that some studies that reported an increase in the CK and LDH levels have been short-term and included more repetitions and sets.

CONCLUSION
Resistance training with blood flow restriction and high-intensity resistance training without blood flow restriction have similar effects on markers of muscle damage. However, more studies are required on the safety and effect of these types of exercise training at different intensities and cuff pressures on other indicators such as myoglobin.

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REFERENCES
1. Correia-Oliveira CR, Lopes-Silva JP, Bertuzzi R, Mcconell GK, Bishop DJ, Lima-Silva AE, Kiss MA. Acidosis, but not alkalosis, affects anaerobic metabolism and performance in a 4-km time trial. Medicine & Science in Sports & Exercise. 2017; 49(9):1899-910.

2. McGinley C, Bishop DJ. Influence of training intensity on adaptations in acid/base transport proteins, muscle buffering capacity, and repeated-sprint ability in active men. J Appl Physiol. 2016; 121: 1290-305.

3. Choung BY, Byun SJ, Suh JG, & Kim TY. Extracellular superoxide dismutase tissue distribution and the patterns of superoxide dismutase mRNA expression following ultraviolet irradiation on mouse skin. Experimental dermatology. 2004; 13(11): 691-99.

4. Cannon J, Marino F.E. Early phase neuromuscular adaptations to high- and low-volume resistance training in untrained young and older women. J Sports Sciences. 2010; 28(14):1505-14.

5. Winett RA. Meta-analyses do not support performance of multiple sets or higher volume resistance training. J Exerc Physiol. 2004; 7 (5): 10-20.

6. Cook S BB, Clark C, Ploutz-Snyder LL. Effects of exercise load and blood-flow restriction on skeletal muscle function. Med Sci Sports Exerc. 2007; 39 (10): 1708-13.

7. Fujita T, Brechue WF, Kurita K, Sato Y, Abe T. Increased muscle volume and strength following six days of low–intensity resistance training with restricted muscle blood flow. Int J Kaatsu Training Res. 2008; 4 (1): 1-8.

8. Tartibian B, Derafshi B, Hajizadeh Maleki B, Tofighi A. The Effect of Indomethacin on Biochemical, Functional and Superficial Symptoms of Delayed Onset Muscle Soreness (DOMS) in Untrained Men. Sport Biosciences. 2010; 3: 93 -110. [In Persian].

9. Sahran Y, Sofian A.M, & Saad A. Z. M. Pretreatment serum lactate dehydrogenase (LDH) and serum alkaline phosphatase (ALP) as prognostic factors in patients with osteosarcoma. J Cancer Prev Curr Res. 2018; 9(2): 58-63. [ In Persian]

10. Mohebbi H, Rahmani-Nia F, yar Arabmomeni, A, Riasi A, & Marandi M. The effects of interval training and age on blood lactate (La) levels and lactate dehydrogenase (LDH) activity in male
1. Nieman DC, Henson DA, McAnulty L. Influence of vitamin C supplementation on oxidative and immune changes after an ultramarathon. Journal of Applied Physiology. 2002; 92(5): 1970-77.

2. Abe T, Kearns C F, Sato Y. Muscle size and strength are increased following walk training with restricted venous blood flow from the leg muscle, kaatsu–walk training. J Appl Physiol 2006; 100 (5): 1460-6.

3. Moore Daniel R, Burgomaster Kirsten A, Schofield Lee M, Gibala Martin J, Sale Digby G, Phillips Stuart M. Neuromuscular adaptations in human muscle following low intensity resistance training with vascular occlusion. Eur J Appl Physiol. 2004; 92 (4-5): 399-406.

4. Abe T, Beekley M, Hinata S, Koizumi K, Sato Y. Day to day change in muscle strength and MRI measured skeletal muscle size during 7 days kaatsu resistance training: A case study. Int J Kaatsu Training Res. 2005; 1 (2): 71-6.

5. Andersen L, Magnusson S P, Suetta C. Changes in the human muscle force velocity relationship in response to resistance training and subsequent detraining. J Apply Phys. 2005; 99(1): 87.

6. Liu T, Kuo C, Wang PS. Exercise and testosterone. Adapt Med. 2009; 1(1): 26-31.

7. Madaram H, Sasaki K, Ishii N. Endocrine responses to upper and lower-limb resistance exercises with blood flow restriction. Physiol Hung. 2010; 97(2): 192-200.

8. Takarada Y, Nakamura Y, Aruga S, Onda T, Miyazaki S, Ishii N. Rapid increase in plasma growth hormone after low-intensity resistance exercise with vascular occlusion. J Appl Physiol. 2000; 88 (1): 61-5.

9. Abdelfattah A.E.E, Salem H.H. Effect of occlusion swimming training on physiological biomarkers and swimming performance. World Journal of Sport Sciences 2011; 4(1): 70-5. [In Persian]

10. Foroughi Parandjuny A, Ibrahim M, Changizii M. The effects of resistance exercise on muscle damage and DOMS measures in athlete male students. Research on college sports. 2015; 3(8): 37-52. [In Persian]

11. Pettersson J, Hindorf U, Persson P, Bengtsson T, Malmqvist U, Werkstrom V. Muscular exercise can cause highly pathological liver function tests in healthy men. British journal of clinical pharmacology. 2008; 65(2): 253-59.

12. Choung B.Y, Byun S.J, Suh J.G, & Kim T.Y. Extracellular superoxide dismutase tissue distribution and the patterns of superoxide dismutase mRNA expression following ultraviolet irradiation on mouse skin. Experimental dermatology. 2004; 13(11): 691-99.

13. Hosseinikak A, Zamand P, Khademosharie M. Compare hormonal responses to two types of resistance training and unrestricted blood flow. Journal of sport biosciences. 2015; 7 (3): 391-405. [In Persian]

14. Nobahar, M. The effects of one progressive session exercise in day during a week on some enzymes muscle damage in active girls. 2013; 1(3): 79-84. [In Persian]