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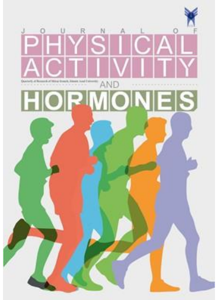

Comparison of two methods of high-intensity aerobic and strength training on serum testosterone, cortisol, insulin-like growth factor1, and immunoglobulin G in professional taekwondo athletes

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Keywords
Testosterone, Cortisol, Insulin-like Growth Hormone (IGF1), Immunoglobulin (IgG), High-intensity Training, Taekwondo Player.

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Abstract

Introduction: The purpose of this study was to determine the difference between two methods of high-intensity aerobic and strength training on serum testosterone, cortisol, insulin-like growth factor1 (IGF1), and immunoglobulin G (IgG) in professional taekwondo athletes.

Material & Methods: This quasi-experimental research was conducted on one group of professional Taekwondo players (n=12) in Rash City with an average age of 22 ± 1.5 years. In three consecutive weeks, three regular training sessions (RTS, 90 minutes), high-intensity strength training (HIST, 80% 1RM, 45min), and high-intensity aerobic training (HIAT, 80% VO2 max, 45 min) were performed. 24 hours after each training session, serum immunoglobulin G (IgG), insulin-like growth hormone (IGF1), plasma testosterone, and cortisol levels were measured after 10 hours of fasting. Data related to the sample are presented with descriptive statistics, and analysis of variance (ANOVA) for repeat measurement was used to evaluate variables. The statistical software program SPSS (SPSS Co, Chicago IL, version 26) was used for data analysis. All statistical tests were performed and considered significant at a $P \leqslant 0.05$.

Results: The findings showed a significant increase in testosterone in both HIAT and HIST groups compared to RTS, cortisol in HIST compared to the other two groups, IGF1 in HIAT compared to the other two groups, but IgG in HIST and HIAT compared to RTS showed a significant decrease ($p < 0.05$).

Conclusion: Based on the research findings, probably both training methods can similarly increase plasma testosterone, HIST increases cortisol, and HIAT increases IGF1 in professional taekwondo athletes, but both HIAT and HIST decrease plasma IgG and this drop is greater in HIAT.

1. Introduction

Participation in sports activities causes acute and chronic metabolic changes through physical training and competition. While the goal of overload training is to create an imbalance between training load and recovery, this overload is only for a short period and for a manageable load that leads to a return to homeostasis. If the load is too stressful or too easy, the overtraining state can be characterized by various normal changes, including molecular, biochemical, and regulatory changes, which may lead to poor performance, well-being, disease, and possible injury (1). When done in moderation, physical exercise can be a stimulant for immune system function. On the other hand, it can have a suppressive effect on the immune system and cause an increase in pro-inflammatory biomarkers during high-intensity training sessions, while when it is performed at a lower intensity, it causes a decrease in the inflammatory state (2).

Growth hormone (GH) is a polypeptide hormone secreted by the anterior pituitary gland and contributes to numerous biological processes such as anabolism, protein synthesis, and substrate movement. Metabolic and anabolic responses associated with growth hormone are mediated through the interaction of the hormone with its receptor, directly by tyrosine kinase activation and indirectly by induction of insulin-like growth factor 1 (IGF-1) (3). All types of exercise induce the release of more growth hormone (GH), and plasma GH levels rise within 10 to 20 minutes after exercise. Insulin-like growth factor-1 (IGF-1), a polypeptide hormone with a homologous structure similar to proinsulin, mediates many of the physiological effects of growth hormone (GH). IGF-1 is synthesized in the liver and secreted into the blood, where it circulates as complexes associated with specific binding proteins (IGFBPs). This set is regulated in two endocrine and tissue-specific and paracrine ways. Studies have shown that IGFBPs may act as transport proteins for IGF-1 to target cells, but can also alter the interaction between IGF-1 and its cellular receptors. Furthermore, IGFBPs may act independently of IGF-I. IGF-1 was recognized as one of the key mediators of carbohydrate, lipid, and protein metabolism, thus maintaining the body's energy balance, which is important for athletes during systematic physical effort. Physical activity can stimulate growth hormone and IGF-1 secretion, and therefore IGF-1 may be related to exercise intensity. Based on the results of studies, the duration and intensity of physical training and the type of training have a significant effect on IGF-1 concentration (4).

Immunoglobulins are a major group of proteins that are an important part of the body's immune system. The main representative of human antibodies is immunoglobulin A (IgA), which is found in saliva, tears, or intestinal mucosa, so it is the first line of defense of the mucosal humoral response. Its functions include the neutralization of toxins and the cleaning of various pathogenic microorganisms, and it also plays an important role in the immunity of the oral mucosa by limiting its colonization and thus preventing the invasion of the epithelium. Serum IgA levels generally decrease after exercise compared to before exercise, and its recovery time is 24 hours. As a result, this immune gap increases susceptibility to upper respiratory tract infections (5).

Studies have shown that high-intensity exercise has a depressing effect on cellular immunity, however, researchers believe that although high-intensity exercise may act as a suppressor for the immune system and increase the risk of respiratory infections, moderate-intensity exercise may it stimulates the defense mechanisms and helps to prevent the occurrence of such pathologies. Improper stimulation and excessive production of catabolic hormones or a decrease in anabolic hormones may lead to an increased risk of musculoskeletal injuries. Hence, the importance of measuring bioenergetics hormones in athletes to prevent some harmful effects caused by excessive training. In the diagnosis of so-called chronic low energy availability (LEA). Changes in serum levels of some endocrine biomarkers can be useful in evaluating available energy. Biochemically relevant decreases in serum levels of insulin-like growth factor-1 (IGF-1), cortisol, thyroid hormones, testosterone, or ovarian hormones have been observed in energy-deficient athletes (6).

In particular, cortisol is a powerful hormone that is controlled by the adrenal cortex and has a catabolic effect. Therefore, an athlete needs to reduce cortisol levels to achieve tissue growth, bone health, ligament health, and positive adaptation to exercise training, while preventing excessive inflammation and tissue damage (7). However, excess cortisol also suppresses the immune system, creating an increased risk for infections and injuries and a concomitant decrease in testosterone levels (1).

Testosterone is an anabolic-androgenic steroid hormone that primarily interacts with androgen receptors in skeletal muscle, whereas the more potent dihydrotestosterone acts primarily in sex-related tissues with a possible secondary role in skeletal muscle. Testosterone exerts many ergogenic, anabolic, and anticatabolic functions in skeletal muscle and neural tissue, resulting in dose-dependent increases in muscle strength, endurance, and hypertrophy (8). At the beginning of the Long-term exercise, testosterone secretion increases and decreases as the action continues. Endocrine adaptation regulates muscle activity, stimulates glycogenolysis, and facilitates gluconeogenesis (9). The studies of Zar et al. (10) have shown that in healthy men, testosterone concentration increases after submaximal and maximal endurance training simultaneously with an increase in cortisol concentration after maximal training and a decrease after submaximal training, while the ratio of testosterone to cortisol It is higher for submaximal exercise compared to maximal exercise.

In any case, maintaining and increasing circulating testosterone concentration along with decreasing cortisol levels can be the main factor in promoting muscle hypertrophy resistance training programs followed by chronic or intense exercise. Evidence suggests that acute resistance training increases testosterone and cortisol concentrations. However, compound activities can have a higher metabolic demand and thus lead to a greater increase in cortisol, which can negatively affect testosterone secretion during a training session. Testosterone and cortisol seem to respond to greater intensity and longer duration of activity. High-intensity exercise protocols that involve large muscles strongly increase testosterone levels, and hormone responses decrease in both sexes when exercise intensity is reduced from 70% to 40% (9).

Taekwondo is one of the intermittent sports in which athletes must train and compete with different intensities using all energy systems. This is because the actions involved in Taekwondo are characterized by periods of high levels of technical, tactical, psychological, physical, and physiological fitness. Therefore, it is important to determine the effect of different training methods, alone or in combination, on blood muscle damage biomarkers (1). Therefore, in the present study, the effect of two methods of high-intensity aerobic and strength training on the serum levels of testosterone, cortisol, insulin-like growth factor-1 (IGF1), and IgG of professional taekwondo athletes is investigated.

2. Materials and methods

2.1. Sample

Twelve professional taekwondo players of the Guilan province team (IRAN) with an average age of 22±1.5 years volunteered to participate in this study. The criteria for inclusion into this study included having a black belt, engagement in intense athletic conditioning for over four years and healthy physical conditions without a history of cardiovascular or endocrine/immune-related diseases. Participants who needed to take any medication during the period of this study were excluded. All participants signed an informed consent form after having been fully informed of the risks and the purpose of the study. The study was reviewed and approved by the Human Ethics Committee of the Rash Branch, Islamic Azad University (IR.IAU.RASHT.REC.1402.033).

This research was conducted for 3 consecutive weeks. All athletes regularly trained three days a week. In the last weekly training session, regular training sessions (RTS, 90 minutes) were performed in the first week, high-intensity strength training (HIST, 80% 1RM, 45min) in the second, and high-intensity aerobic training (HIAT, 80% VO2 max, 45 min) in the third week. (Table1). 24 hours after each three training session, a blood sample was taken from the brachial vein of the right arm. After 12 minutes of centrifugation at a speed of 3000 rpm, the plasma was separated and stored at -280C.

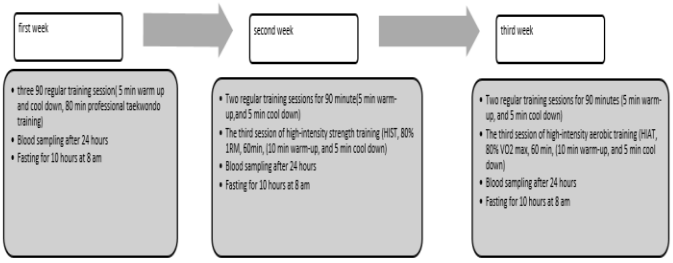

Fig 1. *Training sessions*

2.2. High-intensity aerobic training (HIAT)

The protocol of one session HAIT program included 60 minutes of exercise (10 minutes of stretching and aerobic running with an intensity of 55-60% of maximum heart rate for warm-up (Formula 1), then according to (Table 2), exercise was implemented with an intensity of 80% of VO2max and finally ended with 5 minutes of cooling down by walking slowly on a treadmill (11).

Maximum heart rate = 220-Age

Table 1. *High-intensity aerobic training (HIAT) protocol (Ferguson et al. 2014)*

kilometers per hour	intervention	Time
3-4	Stretching and warming up	10 min
10-12	Speed	1min
7-9	Recovery	2min
10-12	Speed	1min
7-9	Recovery	2min
10-12	Speed	1min
7-9	Recovery	2min
10-12	Speed	1min
7-9	Recovery	2min
10-12	Speed	1min
7-9	Recovery	2min
10-12	Speed	1min
7-9	Recovery	2min
10-12	Speed	1min
7-9	Recovery	2min
10-12	Speed	1min
7-9	Recovery	2min
3-4	Cool down	10 min

2.3. High-intensity strength training (HIST) protocol

One session of the HIST program included 60 minutes of exercise (10 minutes of stretching and aerobic running with an intensity of 55-60% of maximum heart rate for warm-up), then according to Table 3, seven movements included (lower limb strength training, upper limb strength training, balance training) with 5 repetitions in 3 sets and 9 minutes of rest between sets were performed. There was a 3 minute rest between each exercise and a 5-minute cool-down (1).

Table 2- *High intensity strength training protocol*

Type of exercise	Intervention	duration of each move	rest period between sets
Walking and stretching	-	10 min	-
-	3 sets of lower limb strength training with 5 repetitions	6 second	1min
Rest	-	3min	-
-	3 sets of upper limb strength training with 5 repetitions	6 second	1min
Rest	-	3min	-
-	3 sets of balance strength training with 5 repetitions	6 second	1min
Cool	-	5 min	-

2.4. Anthropometric measurements

A Bürer digital scale (Germany) was used to measure the weight. Formula 2 was used to determine body mass index (BMI). The satiety equation (Formula 3) was used to calculate body fat percentage.

$$BMI = \frac{weight}{height^2}$$

$$Body\ fat\ percentage = \frac{495}{BMI} - 450$$

2.5. Maximum repetition (1RM) and maximum oxygen consumption (VO2max)

Before starting the high-intensity strength training protocol, the subjects were familiarized with the equipment and also with the correct techniques of high-intensity strength training. Familiarization with the high-intensity aerobic training protocol started with a 10-minute warm-up of running on a treadmill and performing 8 repetitions of movements with 50% of the individual's expected VO2 max. After 1 minute of rest, the movements were performed for 3 repetitions at 70% of the expected VO2 max. Following high-intensity strength training with 3 minutes of rest, subjects performed 1RM for 3 to 5 attempts and 3 to 5-minute rest intervals with a gradual increase in load or weight (approximately 5%) (12). Brzycki Equation (5) was used to calculate the strength (1RM) in different stages of the test and training.

$$VO2max = 15 \times \frac{HRmax}{HRrest}$$

$$1RM\ (kg) = \frac{(kg)weight}{1 - (0/02 \times Repeat)}$$

2.6. Blood sampling and evaluation of hormone levels

24 hours after the last session of specialized Taekwondo training, after HIAT, and HIST, blood samples were taken after 10-hour fasting. Blood samples were taken from the right-hand antecubital vein. The blood samples were then centrifuged for 12 minutes at a speed of 3000 rpm, and

the serum was separated and stored at -28°C to measure the desired factors.

Testosterone and Cortisol are evaluated by the immunoassay method, (GMBH kit, Germany) with a coefficient of variation of 1.5% and 4.3% respectivly; IGF1 is analyzed by ELISA method, (stat Fax 303 plus, Elisa Awareness, Technology CORP, Palm City, USA), with a coefficient of variation of 6%; Nephrometric method is used to evaluate serum immunoglobulin G (IgG) levels.

2.7. Statistical Methods

Mean and standard deviation indicators were used to provide descriptive statistics. After checking the normal distribution of the data by the Shapiro-Wilk test, repeated measurements of the analysis of variance test (ANOVA) were used. The software used was SPSS version 26 and the significance level was considered as $p<0.05$.

3. Results

The results of the Shapiro-Wilk test showed that the variables have a normal distribution. The characteristics of the research subjects including height, weight, age, and body mass index were described in Table 3.

Table 3- *Demographic characteristic*

Measurement index	mean ± standard deviation
age (years)	22±1.5
height (cm)	173.± 3
weight (kg)	54±7.2
body mass index (BMI)kg/m^2	20.7±1.01
Body fat percentage (kg/cm)	9.2±1.1

Analysis of variance tests with repeated measurements, the research hypothesis has been tested. Table (4) shows the descriptive indices of serum testosterone of professional Taekwondo athletes in three stages: pre-test, post-test of high-intensity aerobic training and post-test of high-intensity strength training.

Table 4- *Serum levels of study variable*

Variable	Regular exercise training	HIST	HIAT
Testosterone (ng/ml)	19.52±3.5	20.81±6.9	20.15±8.3
Cortisol (ng/ml)	16.32±3.9	16.47±3.59	15.90±4.1
IGF1 (ng/ml)	316.84±61	300.96±71.10	325.20±85.96
IgG (mg/dl)	1256.92±262.80	1143.17±213.87	1070.50±260.26

HIST: High-intensity resistance exercise, HIAT: High-intensity aerobic training

The results of repeated measurements of ANOVA were shown in Table 5.

Table 5- The *results of repeated measurement ANOVA*

Variable	Source		sum of squares	F	Sig
Testosterone	Intergroup	Practice	2.407	0.188	0.673
		Practice × group	7.605	0.886	0.367
	between groups	group	14628.9	117.898	0.001*
Cortisol	Intergroup	Practice	1.084	0.245	0.631
		Practice × group	1.003	0.190	0.672
	between groups	group	9483.51	270.450	0.001*
IgF1	Intergroup	Practice	419.170	0.076	0.787
		Practice × group	3220.03	1.849	0.201
	between groups	group	3556996	397.823	0.001*
Ig G	Intergroup	Practice	208507.04	44.008	0.001*
		Practice × group	3375.6	1.560	0.238
	between groups	group	48179794.6	274.315	0.001*

4. Discussion

Based on the results, it was shown that both aerobic and strength training with high intensity can similarly increase plasma testosterone in taekwondo players. Strength training increases testosterone concentration and aerobic training can decrease testosterone concentration. High-intensity training changes the balance of anabolic and catabolic hormones and probably reduces the strength of high-intensity training due to the increase in muscle hypertrophy. Kramer et al. (8) stated that performing high-intensity exercise can change the ratio of testosterone to cortisol and lead a person to the process of anabolism. The aerobic part of training can cause the superiority of the catabolic system, which can limit the development of muscle strength. Testosterone is circulating in the blood in combination with sex hormone globulin or albumin. Due to its high molecular weight, this complex cannot pass through the endothelium of the capillaries, and it is also not possible to penetrate the plasma wall of the cell nucleus to react with nerve agents and regulate their function. For this reason, testosterone probably has a homeostatic dynamic response pattern in high-intensity strength training, which, by converting testosterone into a free form with a lower molecular weight. The increase of free testosterone in the plasma confirms the active homeostatic response in men. Also, increasing total and free testosterone at rest probably causes muscle hypertrophy. In line with the results of the present study, Ganduzi et al. (13) stated that cortisol response and the testosterone/cortisol ratio are related to a victorious fight in combat sports. Sholi et al. (9) found that training at different intensities leads to significant changes in testosterone levels. However, inconsistent with the current research, Hejazi and Hosseini (14) found that the intangibility of testosterone levels did not change in semi-endurance elite runners. This can be due to the difference in the sample characteristics and the type of exercise.

It was shown that there is a significant difference between the two methods of high-intensity aerobic and strength training on the cortisol levels of professional taekwondo athletes. Exercise can lead to higher levels of cortisol in the final stages. Exercise programs that cause an increase in cortisol response can lead to a greater acute response of lactate and growth hormone. In addition, the acute increase of serum cortisol is largely related to serum creatine kinase concentrations. Intense exercise programs that impose a high metabolic load on the body, such as high-volume, moderate-to-high-intensity exercise with short rest periods, lead to the greatest acute response of lactate, cortisol, and small changes during exercise. Strength training with high intensity sometimes reduces the concentration of cortisol during rest time and sometimes does not change it. It seems that increasing the volume of training causes a decrease in endurance performance as a result of decreasing the secretion of catecholamines and sympathetic activity. It has been reported that the increase in cortisol secretion depends on the training status of people, and exercising with high intensity increases its secretion. A decrease in the ratio of testosterone to cortisol is considered one of the signs of high training volume and fatigue in athletes. This decrease can be caused either by an increase in cortisol or by a decrease in testosterone. Garhami et al. (2) reported that the resting levels of cortisol in the strength-endurance group with a recovery interval of eight hours had a significant decrease. Ansari et al. (15) stated that six weeks of aerobic exercise increases the cortisol level in active women, Gandouzi et al. (13) found a significant difference for cortisol and the testosterone/cortisol ratio after the fight. Sholi et al. (9) reported exercise intensity as an independent variable affecting changes in testosterone and cortisol levels. Hejazi and Hosseini (14) showed that cortisol increases significantly after the preparation phase of exercise. Sajedi et al. showed that aerobic and resistance concurrent training was no significantly difference in cortisol levels(16).

It was shown that there is a significant difference between the two methods of high-intensity aerobic and strength training on the serum levels of IGF1 of professional taekwondo athletes. Intense physical activity causes oxidative stress. Oxidative stress also subsequently causes fat peroxidation. In response to endurance activity, oxygen consumption in the human body is systemically multiplied. In the muscles, during exercise, the body faces many demands that cause many physiological changes. In such a situation, homeostasis must be kept constant for life to continue. In this regard, the nervous system and the endocrine system (hormone) in a coordinated action involve movement and all the systems to start and control the physiological processes. The hormonal system is responsible for various metabolic functions of the body, such as the synthesis and activation of cell enzymes, changes in cell membrane permeability, protein synthesis, changes in cell metabolism, and stimulation of cell secretion and growth. It is believed that the production of an intermediary protein in the liver and other cells called IGF-1 is exerted on the body. Habibian and Valinejad (17) showed that while no significant difference was observed between the response of IGF-1 to different intensities of endurance exercise, this discrepancy can be attributed to the type of exercise that was only endurance. Ye et al. (18) also showed in line with the present study that strength training can increase insulin-like growth factor 1, and finally, Choobineh et al. (19) There was no significant difference between the training groups in GH and IGF-1 levels. Khosravi et al. showed that concurrent resistance training (75–85% of 1RM) immediately followed by endurance training for 30 minutes using an ergometer (75-85% of HRmax) did not affect the response of cortisol, testosterone, insulin, and insulin-like growth factor (IGF-1) to athlete's women(20).

This study results showed that HIST increases plasma cortisol, while HIST and HIAT compared to RTS showed a significant decrease in the serum levels of IgG of professional taekwondo athletes. It can be said that high-intensity exercise may suppress the immune system for 1 to 6 hours of recovery. According to the "j-loop" hypothesis, high-intensity exercise may increase the risk of upper respiratory tract infections. For example, an intense exercise session may suppress the innate immune system for up to 24 hours after exercise. During this "open window" the risk of disease may increase (McFarlane et al., 2005). The binding of Ig to its target antigen forms antibody-antigen complexes. The effect of exercise on humoral immune function has been assessed by measuring serum and mucosal Ig concentrations in vivo and serum immunoglobulin (S-Ig) synthesis following in vitro mitogen stimulation (4). Arshadi et al. (21) showed that performing maximal anaerobic activity following sleep and insomnia causes a significant increase in cortisol and IgA. The variables and controls were measured, especially in the sleep regulation section, but Jalili et al. (22) showed that serum IgA did not change much before and after two hours after training. Among the limitations of this research, we can mention the lack of diet control and the level of anxiety of the samples, which affects the hormones controlled in this research.

5. Conclusion

Based on the research, it was shown that there may be a significant difference between the two methods of high-intensity aerobic and strength training on the testosterone serum levels of professional taekwondo athletes. It has been observed that testosterone may increase after both HIST and HIAT. Also, it was shown that there may be a significant difference between the two methods of high-intensity aerobic and strength training on the cortisol levels of professional taekwondo athletes. Cortisol levels may increase after a high-intensity strength training test. On the other hand, it was shown that there may be a significant difference between the two methods of high-intensity aerobic and strength training on the serum levels of IGF of professional taekwondo athletes. It has been observed that the serum levels IGF1 may increase after the HIAT. It was shown that there may be a significant decrease the two methods of aerobic and strength training with high intensity on the serum levels of IgG of professional taekwondo athletes.

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Conflict of interests: The authors declare that they have no conflict of interest relating to the publication of this manuscript.

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