

Effect of the Low or High Volume of High-Intensity Interval Training Protocols on the Leptin and Lipid Profile in Men with Type 2 Diabetes

Hamidreza Malekinezhad¹ , Dariush Moflehi² , Hamid Abbasi^{*3} , Arezoo Behzadi¹ 

1. Department of Physical Education and Sport Sciences, Shahid Bahonar University of Kerman, Kerman, Iran
2. Department of Sport Physiology, Faculty of Physical Education and Sport Sciences, Shahid Bahonar University of Kerman, Kerman Iran
3. Department of Sport Sciences, Faculty of Psychology, Yazd University, Yazd, Iran

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Corresponding Author:

Hamid Abbasi
habbassi@yazd.ac.ir

ABSTRACT

Introduction: Sport activity leads to metabolic and physiological adaptations, which may have a beneficial role for diabetic patients. The purpose of this study was to compare the effects of low volume and high volume high-intensity interval training (HIIT) on serum leptin and some of the lipid profiles of type 2 diabetic men.

Methods: In this randomized clinical trial, 30 males with type 2 diabetes mellitus were divided into three groups of 10, including low volume and high volume HIIT and control groups. The low volume groups underwent intensive training for up to two minutes and a high volume group up to six minutes for 8 weeks. Blood lipid profile indicators were measured before and 48 hours after the last exercise session. By using SPSS version 25, the paired t-test, ANOVA, ANCOVA and LSD as a post hoc test were used to analyze the data. For all analysis $\alpha=0.05$ was considered as the level of statistical significance.

Results: There was a significant difference before and after 8 weeks of low-volume HIIT training at triglyceride ($p = 0.049$). However, there was no significant difference in serum leptin, cholesterol, LDL-C, and HDL-C in men with type 2 diabetes ($p>0.05$). ANCOVA only found a differences between groups on BMI ($F_{2, 26} = 3.61, p = 0.041$). Also, the post hoc in two by two comparisons of HDL showed that there was a significant difference between high volume HIIT and control groups ($p = 0.021$) and low volume and control group ($p = 0.011$). The HDL-C levels in HIIT training groups increased significantly compared to controls.

Discussion: Considering other proven adaptations of HIIT training on triglyceride, HDL and improving glycemic indexes is a useful and safe training for type 2 diabetic patients. Therefore, needing a very short time for this training, metabolic, functional and physiological adaptations of HIIT training, along with other exercises are recommended to improve the condition and reduce the risk of cardiovascular disease and complications of type 2 diabetes.

Keywords: Leptin, High-intensity Interval training, Diabetes mellitus, type 2, Lipids profile

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Introduction

Type 2 diabetes is a common chronic disease that is associated with insulin resistance and increased blood glucose (1). Type 2 diabetes is one of the life-threatening metabolic disturbances. WHO in 2019 reported that the prevalence of diabetes has been steadily increasing for the past 3 decades about 422 million people worldwide have diabetes (2) and it has been estimated that around four million adults in Iran have Type II diabetes in 2014, and the main problem for diabetic patients who have a problem with the prevention and control of this disease is lack of awareness and attention to the illness (3). The features of this metabolic disorder are hyperglycemia, impaired metabolism of carbohydrates, fats, and proteins, along with a complete or partial deficiency of insulin secretion in the body (4). Former studies have shown that the risk of coronary heart disease in type 2 diabetic patients is 2 to 4 times more than the normal people (5). Insulin resistance and impaired insulin function can lead to dyslipidemia in diabetic patients. Increasing the concentration of triglyceride, LDL cholesterol and lowering the HDL cholesterol concentration are dyslipidemic properties in diabetic patients, which are also important risk factors for atherosclerosis and cardiovascular diseases (6). Diabetes mellitus in addition to dyslipidemia causes a large, quantitative and qualitative change in the types of lipids and lipoproteins (7). High levels of leptin (leptin resistance) are associated with high levels of obesity, glucose intolerance, excessive triglyceride levels, and high blood pressure, which is the cause of the metabolic syndrome. Such metabolic disorders can ultimately lead to heart disease, stroke, and type 2 diabetes (8). On the other hand, despite the fact that the relationship between leptin and insulin is not completely clear, some reports suggest the effects of leptin on blood glucose as an anti-diabetic agent (9). This hormone acts as a fat mass sensor, and its concentration in the blood is strongly related to body fat stores and obesity. Studies have shown that leptin enhances the absorption of glucose and fatty acids, as well as increasing the oxidation of fatty acids in skeletal

muscle, as well as decreasing the content of triglycerides, increasing the withdrawal and oxidation of fatty acids in the liver tissue (10). Leptin, directly or through the activation of certain centers in the central nervous system, reduces the intake of food, increases energy consumption, affects on glucose and fat metabolism, and changes in neuroendocrine activity. Leptin receptors exist in pancreatic beta cells, and play an important role in the secretion of insulin from these cells. Increasing blood glucose in the presence of high insulin and leptin levels in individuals points to the resistance of these individuals to these two hormones (8).

Regarding the effect of exercise on insulin resistance and leptin hormone, some studies have shown the positive effects of traditional aerobic and resistant and combined training on these factors (11 and 12), while some studies also have shown no significant effect of these exercises on the levels of serum leptin (13 and 14), recently, HIIT exercises have been considered by sports and clinical researchers for its optimal systemic, cellular and metabolic compatibility in terms of time. However, research into the effects of these exercises on diabetes is still in the early stages of the study, and the results of studies about the effect of these exercises on various aspects of diabetes, especially biochemical changes in adipose tissue and lipid profiles are contradictory. Former studies showed high-intensity interval training has a significant effect on serum leptin decrease (15-18), while other studies couldn't find any significant effects on leptin (19, 20). Given that these interventions were different in terms of exercise protocols and in terms of severity, duration, and training periods, it can be said that the effect of HIIT exercises on serum leptin levels depends on the severity, duration, and training period. The contradictions in the effects of HIIT exercises on serum leptin and lipid profiles, based on the literature, the intensity and duration of HIIT exercises, have a significant impact on the effects of different types of HIIT exercises. Therefore, the researchers in this study, given the limited research

background in the comparison of the effects of low-volume and high-volume protocols of this type of exercise on diabetic patients, and in particular the levels of leptin and lipid profiles, which are among the factors affecting cardiac pathologies, the vascularity of these patients and their association with complications of diabetes mellitus are sought after to respond to the impact of high and low HIIT exercises as an optimal training method on serum leptin levels and lipid profiles in men with type 2 diabetes.

Methods

The present study was a randomized control trial study with a pretest and post-test design with the control group. The statistical population of this study included all non-athletic type II diabetic men in Yazd. The sample size was calculated by using G*power software based on the mean and standard deviation of previous studies for blood glucose, insulin and insulin resistance (15). The minimum required number of 9 samples for each group was estimated ($\alpha = 0.05$, power = 0.8 and effect size = 0.25), which for a higher statistical power of 10 samples per group was chosen. Subjects consisting of 30 patients with type II diabetes mellitus who were refer to diabetes research center of Yazd medical university were selected and by using simple random sampling were divided into three groups: high volume HIIT training (n=10 subjects), low volume HIIT (n = 10) and the control group (n = 10). During the study, the training groups participated in the HIIT training protocols for an extended period of 8 weeks, and the control group did not receive any interventions during this period, and only had their daily and normal activities. Subjects were evaluated for physical health, history of cardiovascular, respiratory, orthopedic problems and diabetic neuropathy under the supervision of physicians before the intervention. Then, the purpose and methods of performing the work were explained to the subjects during the first session. All subjects completed the PAR-Q questionnaire and medical assessment and signed the informed consent. Subjects were non-athletic and non-insulin

dependent with an age range of 29 to 46 years and BMI range of 23 to 36. The severity of the disease in these subjects was in terms of fasting glucose levels from 126 to 350, after two-hour glucose level from 200 to 400, and HbA1c higher than 6.5 percent.

It should be noted that the subjects in this study used metformin and glibenclamide drugs under the supervision of their physician. All specimens had a minimum FBS of 126 and a two-hour glucose test of 200 (Glucose Tolerance Test) and HbA1C of greater than 6.5 for inclusion in the study.

The variables that have been measured in this study were leptin and lipid profile including cholesterol, triglycerides, HDL, LDL.

In each group, all of the dependent variables of the research were collected after signing informed consent and completion of relevant forms, such as PAR-Q, perform exercise test by the physician 24 hours prior to the beginning of the training. The exercise as the interventions was performed in the sport club of diabetes research center of Yazd.

To estimate insulin resistance, a hemostasis model was used to measure fasting insulin levels in fasting glucose levels based on 22.5%. The serum level of lipid variables was measured by an enzymatic method using specific kits of triglyceride, cholesterol, HDL and LDL from Biosystems which is an Italian company. The groups were matched based on the body mass index. The exercises on the Chinese classic model bike that was made in China and the Polar chassis pulse watch (Polar Equine S-610i, Polar®) was used to control the intensity of exercise during exercise. In order to prevent blood sugar loss during and after exercise, 200 ml of juice was available per person per session.

Accordingly, the training program included 2 Protocols HIIT exercises, low and voluminous which were performed for eight weeks and three non-consecutive days each week. Each training session consisted of five minutes of warming (stretching and walking on a stationary bike) and five minutes of cooling (very low intensity). The first two weeks included lower intensity exercises for compatibility and training as well as training.

Low-volume HIIT training protocol

The training program included 24 sessions for 8 weeks and 3 non- consecutive days each week. The exercises began in the first six sessions to create adaptability and training with very low intensity. The repetition of the training sets from the fifth session to the next changed from the 2 sets to the 6 sets in the last session. The duration of each set was from 10 seconds to 20 seconds. In order to comply with the principle of overload, the intensity of exercise was increased by using heart rate.

High-volume HIIT exercise protocol

The training program included 24 sessions for 8 weeks and 3 non- consecutive days each week. The exercises began in the first six sessions to create adaptability and training with very low intensity. The repetition of the training sets changed from 1 to 6 sets in the last session and the duration of each

session was unchanged until the last session that was 60 seconds. In order to comply with the principle of overload, the intensity of exercise was increased by using heart rate.

Statistical analysis

The Shapiro Wilk test was used to evaluate the normal distribution of data. The paired t-test was used to determine the effect of training by comparing the pre-test and post-test values, and for the comparison of the groups, the one way ANOVA and ANCOVA were used and the LSD post hoc test was used to compare the two by two comparisons. All statistical operations were performed using SPSS version 23 software, and Excel 2016 software was used for the graphs. The significance level for the whole hypothesis test was considered 0.05.

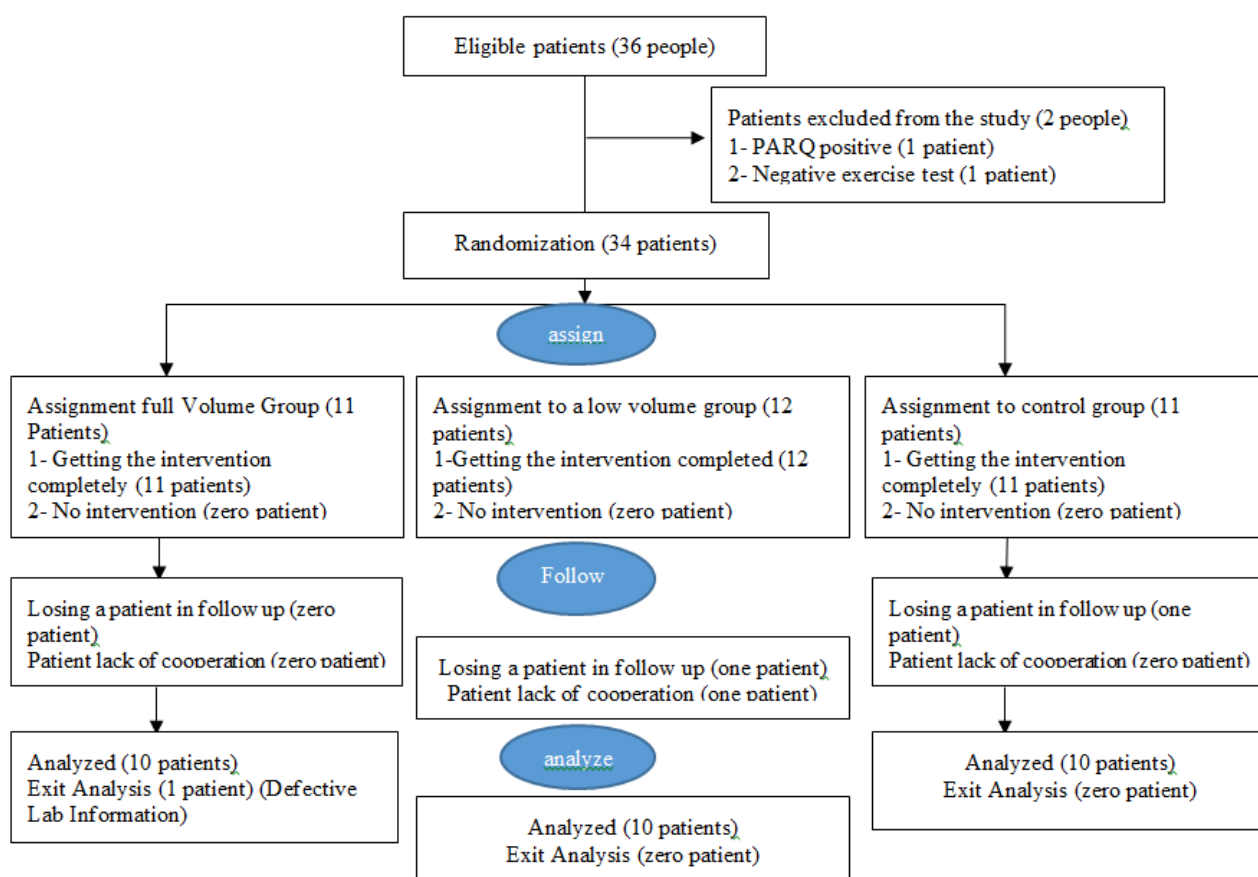


Figure 1. (CONSORT format) Study flowchart

Results

In general, 36 patients were included in this study; six patients were dropped out and only 30 patients were analyzed (10 patients in each group: control, high volume and low volume groups). All subjects were male and the mean age of them was 38.2 ± 4.84 years (range 29-46 years). In the present study, before the intervention there were no difference between groups in age ($p = 0.688$),

BMI ($p = 0.97$) and abdominal circumference ($p = 0.936$).

Also, there was no significant difference in triglyceride, cholesterol, HDL, and leptin ($p < 0.05$) between the three groups in both times before and after the intervention (Table 1). However, LDL before and after the intervention had the highest level in the high volume group and had the lowest level in the control group ($p < 0.05$).

Table 1. Mean and standard deviation of the variables studied in three groups, control, low volume and high volume groups

Variable		Control (n = 10)	High volume (n = 10)	Low volume (n = 10)	P-values ^a
Age (years)		37.5 ±5.78	37.80±3.77	39.30 ±5.10	0.688
BMI (kg.m2)	pre	29.51 ±3.27	29.58±3.14	29.22 ±3.80	0.97
	Post	29.44 ±3.48	29.62±3.36	28.65 ±3.44	0.797
TGL (mg.dL)	Pre	241.20±94.35	213.80 ±81.25	237.80±75.91	0.837
	Post	299.0 ±82.07	194.80±65.16	202.90±80.64	0.441
Coolestrol (mg.dL)	Pre	158.40±26.16	179.80±29.41	180±35.40	0.208
	Post	158.30±23.75	181.50*±18.20	173.60±32.16	0.136
LDL (mg.dL)	Pre	66.90±16.59	97.90* ±26.03	89.80* ±21	0.012
	Post	78.50 ±16.22	105.20*±21.38	87.20*±18.64	0.019
HDL (mg.dL)	Pre	37.90±2.64	39.20*±4.52	39 ±4.76	0.517
	Post	33.10±3.48	37.70±5.77	38.50±2.06	0.056
Leptin (µg.ml)	Pre	8.42±5.92	9.39±4.78	11.31±8.12	0.624
	Post	7.96±4.64	9.91 ±4.51	9.76±7.36	0.761

^a Groups differences examined by one-way ANOVA in pretest and posttest
 * significant differences by the control group (post hoc: LSD)

In the control group, the paired T-test showed that there was no significant difference in triglyceride, cholesterol, LDL, and leptin levels ($p > 0.05$) but, there was a significant reduction (8.4-unit reduction) in HDL level in this group ($p = 0.007$). In the other two groups (low volume and volume groups), the changes in triglyceride were only significant ($p = 0.049$) and there were no significant changes in cholesterol, LDL, HDL, and leptin levels ($p > 0.05$).

The ANCOVA test showed pretest score had an impact on posttest score in all variable except cholesterol, but after removing effect of pretest score, there was a significant difference between the groups on the posttest only for BMI ($F_{2,26} = 3.61, p = 0.041$) and post hoc showed there was a significant difference between the low

volume group and high volume group ($p = 0.018$) and low volume group and control was borderline, not significant ($p = 0.051$).

In terms of HDL, a borderline but not a significant difference between the groups ($F_{2,26} = 3.03, p = 0.056$) was found and in two by two comparison by LSD, there was a significant difference between the high volume group and control group ($p = 0.021$) and control volume ($p = 0.011$).

The cholesterol, triglyceride, LDL cholesterol, and leptin did not show any significant difference between the groups after removing the pre-test score ($p > 0.05$). For post hoc, the LSD showed there was no significant difference between the groups ($p < 0.05$).

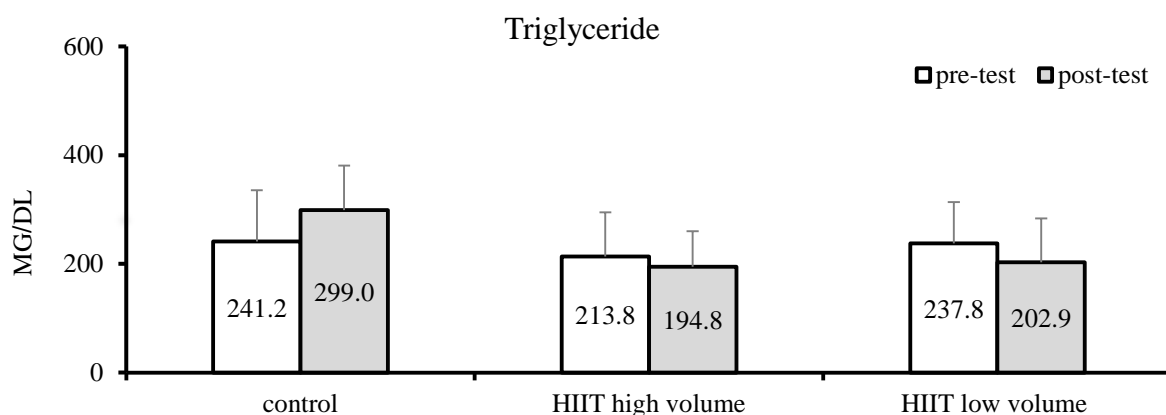


Figure 2. The mean of triglyceride before and after test in all three groups: control, high and low volume HIIT

Discussion

In this study, it was found that except the triglyceride ($p = 0.049$), there was no significant difference before and after 8 weeks of low volume and high volume HIIT exercises in serum leptin, cholesterol, LDL-C and HDL-C in type 2 diabetic men. By comparing two by two groups, between high volume and control groups and low volume and control, HDL-C levels in training groups were significantly higher than the control group.

One of the important components of lipid profiles is the measurement of triglyceride levels in the blood. Insulin resistance and impaired insulin function can lead to dyslipidemia in diabetic patients. Increasing the concentration of triglyceride, LDL cholesterol and lowering the HDL cholesterol concentration are dyslipidemic properties in patients with type 2 diabetes, which are also important risk factors for atherosclerosis and cardiovascular disease. Some studies have also suggested triglyceride as an independent risk factor for heart attacks. Researchers have described the time and period of exercise training as one of the factors influencing lipid profile changes (21). On the other hand, literature has supported the role of dietary supplements alongside exercise in reducing the levels of serum triglyceride (22, 23). However, in this study, due to the lack of proper control of subjects' nutrition, this confounding factor can be considered as one of the reasons for not observing a significant change in other lipid factors. Also, the exercise protocols used in terms of body muscle

involvement (ergometer bicycle) and lack of significant metabolic stress due to the involvement of energy-dependent energy systems can be one of the other reasons for not significantly reducing serum cholesterol and LDL levels. At the same time, recent suggestions suggest that lipid-lowering responses such as TG to exercise are an acute phenomenon that would be lost almost immediately after exercise interruptions, even in highly-skilled individuals. This is a message for researchers and practitioners in the field of physical exercise physiology that exercises should be conducted regularly in order to repeat and maintain their beneficial effects (22, 23). In sum, the results of this study are consistent with the results of Gharari et al. (2014) (22). This research showed that the short duration of the training period (6 weeks of HIIT) is one of the factors affecting the lack of a significant decrease in serum lipid profiles in the subjects. It also suggested that six weeks of HIIT led to an inappropriate improvement in the lipid profiles in passive young men. Whyte et al. (2010) (21) examined the effect of two weeks of speed interval training (SIT) on lipid profiles and health-related factors in overweight and inactive men. The training included four to six 30-second repeat Wingate tests and 270 seconds of rest between each repetition. The results showed that the SIT improved some of the variables associated with cardiovascular disease. This change was not significant. They did not change the lipid profiles for a long time. This

study was consistent with the results of the present study.

Exercise affects the lipid profiles of individuals who have lower levels of TG, LDL-C, or lower HDL-C (22). In this study, the levels of resting LDL-C and HDL-C in the desirable range in all three groups were pre-test, which can be one of the reasons that exercise didn't have an effect on LDL and HDL. In addition to this, researchers point out that exercise training is one of the factors influencing lipid profile changes (21). It has been argued that the beneficial effects of exercise on different lipoproteins are highly correlated with high-intensity exercises. The volume and intensity of the training result in a greater improvement compared to the lower volume and intensity (24). It has been reported that at least eight weeks of aerobic or anaerobic routine training are required to improve HDL-C, and in studies, less than eight weeks, no changes in lipid profiles have been reported. However, there are only few studies which supported blood lipid variables changes after 8 weeks of exercises (22).

Based on the literature, leptin administration to insulin-resistant mice has reduced their resistance. In humans, insulin resistance is associated with high levels of leptin, and many studies have similar levels of leptin in patients with type 2 diabetes, so leptin can also be involved in the pathogenesis of these diseases. One of the regulators of leptin secretion is exercise stress, the concentration of systemic hormones, and the amount of energy consumed. Reducing fat mass is one of the reasons for changing levels of leptin (16, 17). Although some studies have shown that short-term exercise can not affect leptin secretion, short-term changes due to exercise activity in energy balance affect leptin secretion at night. Negative energy balance which is caused by physical activity or reduced energy intake suppresses leptin secretion overnight. However, positive energy balance leads to increased leptin secretion (25). Physical activity is the most important factor in human energy consumption. Since energy can regulate leptin gene expression positively or negatively, the change in energy consumption through exercise

may also affect leptin levels (17, 25). The stress of exercise a strong and specific regulator of leptin. Physical activities have an important role in changes in the flow of fuel in circulating hormonal concentrations and in energy consumption due to physical activities can lead to changes in leptin concentrations (19, 25).

The results of the present study on the effect of intensive interval training on serum leptin levels are inconsistent with the results of the other studies such as Racil et al. (12 weeks training) (2016) (15), Avazpoor et al. (8 weeks) (2016) (16) Soori et al. (16 weeks training) (2012) (17), Inoue et al. (8 weeks) (2017) (18). All of these studies indicated that high-intensity interval training had a significant effect on leptin. In contrast, the results of this study is consistent with the results of the studies by ZilaeiBouri et al. (8 weeks) (2012) (19), and Kong et al. (5 weeks) (2016) (20) that reported the lack of significant effect of HIIT exercises on leptin. Given that these interventions were different in terms of exercise protocols in terms of intensity, duration, and training periods, it can be said that the effect of HIIT exercises on serum leptin levels depends on intensity, duration, and training period.

Exercise reduces serum leptin levels by evacuating glycogen, inhibiting glycolysis, increasing glucose uptake in the presence of lactate, acidosis, and catecholamines. It can also be said that decreasing the concentration of leptin through exercise is associated with changes in energy balance, insulin sensitivity, changes in hormones associated with carbohydrate metabolism and fat. Therefore, the intensity, frequency and duration of the training program should be appropriate in order to change the level of leptin and its dependent hormones, such as insulin, thyroxine, triiodotaurine and cortisol. Most studies conducted using this type of exercise showed its positive effects on some metabolic indexes. Although, changes in leptin levels in serum have been reported in relation to aerobic exercises, and the correlation between changes in leptin with BMI and WHR. On the other hand, the significant reduction of these two variables in response to HIIT exercises suggests a possible

improvement in serum leptin levels as a result of such exercises with higher energy consumption (25, 26). On the other hand, considering that these interventions were different in terms of exercise protocols in the variables of intensity, duration, and training period, it can be said that the effect of HIIT exercises on serum leptin levels depending on intensity, duration, and training period can be different. The contradictions in the effects of HIIT exercises on serum leptin and lipid profiles, as well as the fact that based on the former studies, both the factors were influenced by the intensity and duration of HIIT exercises, now it is interesting to study the effects of a variety of HIIT exercises belong to different training intensity and duration.

Conclusion

In this study, there was a significant difference before and after 8 weeks of low volume and high volume HIIT training in triglyceride, but in serum leptin, LDL-C and HDL-C in men with type 2 diabetes, there was no significant difference. By two by two comparison of the groups, between the high volume group and the control there was a significant difference between the low volume and the control group, and HDL-C in training groups compared to the control group. Considering other proven adaptations of HIIT training on triglyceride, HDL, and improving cardio-respiratory fitness, muscle fitness and function, and physical fitness, as well as improving glycemic indexes this kind of training includes safe exercises for type 2 diabetic patients. Therefore, considering the time-consuming, metabolic, functional and physiological adaptations of the HIIT exercises, it is recommended that these exercises be used along with other exercises such as

moderate aerobic and resistance training to improve the conditions of the disease and reduce the risk of cardiovascular disease and complications of type 2 diabetes patients.

Limitations

The subjects were men for homogeneity of subjects and the possibility of the same intensity of training and also cultural issues for using the same trainers, and it is suggested to do the same experiment on female subjects for future studies.

In this study, higher sample size was chosen, so that in case of a drop out of subjects, there would be no disruption in the results of the study. Among other constraints, it was not possible to examine other effects on leptin including glycogen discharging, glycolysis inhibition, increased glucose uptake in the presence of lactate, acidosis and catecholamines in the studied patients. Due to the lack of sample volume in the high prevalence of dyslipidemia and diabetes, another study with a higher sample size is needed to study other factors that are affected by HIIT training, so that all the effects of HIIT exercises can be taken into account.

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Conflict of Interests

There is no conflict of interest.

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