



# The Effect of Eight Weeks High Intensity Interval Training (HIIT) on E-Selection and P-Selection in Young Obese Females

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## Abstract

**Objectives:** The purpose of this study was to analyze the effect of eight weeks high intensity interval training on the levels of E-selection and P-selection.

**Methods:** 16 obese young females were randomly assigned into two experimental (N = 8, age; 26.12 ± 3.1 years; height, 1.6 ± 0.06 meters; weight, 85.5 ± 16.6 kilograms) and control groups (N = 8; age, 31 ± 6.23 years; height, 1.6 ± 0.02 meters; weight, 76.2 ± 12.06 kilograms). Exercise protocol included eight weeks (three sessions per week, each session four to seven repetitions of running with maximum velocity in a 40-meter distance with 30 seconds' recovery) HIIT trainings. Fasting blood samples were taken 24h before and after the training. Elisa method was used to measure the dependent variables.

**Results:** Analysis of data using student T-test showed no significant changes in the levels of E-selection and P-selection after trainings (P > 0.05).

**Conclusions:** It was concluded that more duration of training is probably needed to have significant effect of HIIT trainings on the levels of E-selection and P-selection.

**Keywords:** High Intensity Interval Training, E-Selection, P-Selection, Obesity

## 1. Background

One of the main causes of morbidity and mortality throughout the world is coronary artery disease (atherosclerosis) which is increasingly expanding worldwide. Based on evidences, the cardiovascular disease is the leading cause of deaths in Iran which could be due to rapid changes in people lifestyle (1, 2). One of the atherosclerosis risk factors is adhesion molecules, which consists of five main categories of integrin, immunoglobulin, selectins, mucin and cadherin. Selectins are among adhesion receptors and can be found on leukocytes (L-selectin), platelets (P-selectin) and endothelium (E-selectin). Selectins are named due to having a semi-lectins domain. P-selectin is important in the first phase of leukocyte recruitment to thrombosis and induction of fibrin.

Production in hemostasis and E-selectin is the most dedicated of inducer molecule in the endothelium surface which is involved in adhesion of neutrophils, monocytes and T-lymphocytes for areas of inflammation (3). Selectins by facilitating the adhesion of monocytes, neutrophils and lymphocytes to the vascular wall cause their migration to

the place below endothelium. In experimental studies of animal, increased sloping was seen in P-selectin levels after consuming a rich diet of cholesterol and reduces the formation of atherogenic plaque and migration of leukocytes after injection of antibodies against P-selectin. Currently, clinics do not frequently use the selectins as predictive markers of future cardiovascular events. However, the development of treatment strategies to reduce the levels and stopping the function of these substances can be very useful in the prevention of atherosclerosis (4, 5). Numerous researches have conducted about the effect of exercise training on selectins levels, but so far, no research has been conducted on the long-term impact of high intensity interval training (HIIT) on the levels of these molecules in obese women. Hamedinia and Haghghi (2007) examined the effect of 13 weeks of resistance exercises (with the intensity of 50% to 60% of one maximum repetition) and endurance (with the intensity of 75% to 85% of maximum heart rate (HR<sub>max</sub>)) on adhesion molecules in almost obese men. The results suggested that both types of exercises had a significant effect on selectins decrease (4). It was also shown in a study that 20 weeks of exercise training reduced P-selectin

levels in patients with chronic heart failure (6). Boos et al. (2008) showed that Bruce protocol caused the increase of E-selectin (7). Saetre et al. (2011) reported E-selectin reduction following eight weeks of walking workout in patients with peripheral artery disease (8). Based on the evidences, regular physical activity improves endothelial function, blood flow development, lipid profiles and also reduces the risk factors for atherosclerosis (9); while the most effective exercise protocols which have the most preventive and curative benefits have not been determined (10). One of the recent exercises protocols, involving alternating periods of intense activity and rest is HIIT training which improves the cardiovascular fitness more than moderate-intensity continuous exercise, and therefore it has more protective role against cardiovascular disease (11). Recent studies have shown that, the number of morbidity and mortality due to cardiovascular disease in women was more than men since 1984. One possible reason may be due to poor physical activity in women (12).

Given the importance of obesity and sedentary life among women and their relationship to atherosclerosis, we studied the effects of eight weeks of HIIT trainings on E-selectin and P-selectin in obese young female.

## 2. Methods

The current study was a quasi-experimental research with pre and post-test design in experimental and control groups. The statistical population included female students (Islamic Azad University of Bojnourd) who were suffering from obesity. The study samples were selected voluntarily and were divided into experimental ( $n = 8$ ) and control group ( $n = 8$ ). Prior to beginning the study, the subjects were given the consent form and their demographic questionnaire and medical and physical activity records were collected. Inclusive criteria consisted of; obesity (body mass index (BMI)  $28 - 38\text{kg/m}^2$ ), lack of illness history, no regular physical activity during the past six months, nonsmokers and the lack of medication therapy for cholesterol, glucose, blood pressure and lack of musculoskeletal injury as a barrier for doing physical activity.

HIIT program consisted of high-intensity of ( $\geq 90\%$  of maximum heart rate ( $\text{HR}_{\text{max}}$ ) for 8 weeks (three days a week). Each training session was begun by general warm up for 10 minutes (walking, slow running, stretching, rotating and jumping); then HIIT training was done at the intensity of 90 to 95% of maximum heart rate ( $\text{HR}_{\text{max}}$ ) and finally, cooling down phase was performed for 10 minutes (jogging, walking and stretching). Maximum Heart rate ( $\text{HR}_{\text{max}}$ ) of the subjects was calculated by the formula of  $220 - \text{age}$ . The exercise intensity was controlled by Polar rate

monitor (Polar X Trainer Plus) that was made in South Korea. HIITAs as shown in Figure 1, HI.

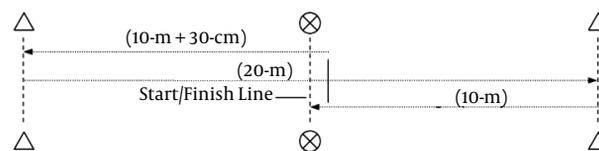


Figure 1. HIIT Protocol

Firstly, the subjects had to pass the routes respectively, 10 m (route 1); then 20 m (route 2) and finally 10 m (route 3) with maximum speed. The total distance was 40 meters that lasted for 30 seconds each time. The protocol was repeated again after a 30 seconds rest. Based on the progressive overload principle, the number of repetitions of 30 seconds in each session was 4 times in the first two weeks, 5 times in the 3rd and 4th weeks, 6 times in 5th and 6th weeks and finally 7 times in the 7th and 8th weeks (13).

All measurements including weight, BMI, cardiovascular endurance of E-selectin and P-selectin levels of subjects were measured before and after HIIT training. Running test was used to determine the level of cardiovascular endurance (14). In a way that subjects ran one mile with their maximum velocity and the time was recorded. To analyze the E-selectin and P-selectin levels, blood samples were taken after 12 hours of fasting in two steps of pre-test and post-test. In pre-test step of taking blood sample, the subjects were asked not to do any hard work two days before the test. Blood samples were taken from the left brachial vein of subjects in a sitting and resting position and samples were immediately sent to the laboratory for analysis by specialist. Blood samples were centrifuged and separated up to an hour after taking the blood samples at 2700 rpm for 10 to 15 minutes. After finishing the exercise period (eight weeks) and 24 hours after the last training session, the blood samples were taken like the first step. The serum was kept in the freezer and at the temperature of  $-20$  degrees centigrade. All the biochemical procedures were performed by auto analyzer machine. All samplings were conducted between 8 to 9 am. ELISA technique was used to determine the amount of E-selectin and P-selectin of serum by using a stat fax / 2100 machine Made in America and Crystal day Science lab kit was made in China.

After data collection, the natural data distribution was analyzed by using the Kolmogorov-Smirnov test and after confirming it, the paired sample t-test was used to evaluate the variation within groups and independent t-test was used to evaluate the variation between groups. All calculations were carried out by using SPSS software version 23 at

the significant level of  $P \leq 0.05$ .

### 3. Results

Table 1 shows the demographic characteristics of subjects (age, height and weight). According to Kolmogorov-Smirnov test the subjects of control and experimental groups were homogenous in values of age, height, and weight before the beginning of the study (Table 2). Data analysis showed that body weight and BMI of the subjects in the experimental group were significantly reduced and the cardiovascular endurance of subjects significantly increased ( $P \leq 0.05$ ); E-selectin and P-selectin levels were reduced not statistically ( $P > 0.05$ ). Paired t-test results are shown in Table 3. As shown in Table 4, all factors including weight, BMI, E-selectin and P-selectin were not statistically changed ( $P > 0.05$ ) except cardiovascular endurance ( $P = 0.001$ ).

Table 1. Demographic Characteristics of the Subjects

Group	Age, y	Height, m	Weight, kg
Experimental	26.1 ± 3.1	1.6 ± 0.06	85.5 ± 16.6
Control	31 ± 6.2	1.6 ± 0.02	76.2 ± 12.06

Table 2. Kolmogorov-Smirnov Test Results

KS Test	Age, y	Height, m	Weight, kg
Z-score	0.17	0.13	0.18
P value	0.19	0.2	0.13

### 4. Discussion

The results suggested that weight and BMI were reduced after eight weeks of HIIT training while the cardiovascular endurance was increased. These findings were consistent with Roan et al. (2012), Soori et al. (2012), Kordi et al. (2013) and Khodaei et al. (2014) (15-19). Roan et al. (2012) which suggested that five weeks of HIIT training increased the cardiovascular endurance in female soccer players. They stated that two training sessions a week of HIIT can be a good stimulus for increasing cardiovascular endurance (15). In Sandvei et al. (2012) study, eight weeks of speed interval training which is similar to HIIT training, increased the cardiovascular endurance in healthy young men and women, but they had no significant effect on weight and BMI (16). Soori et al. (2012) studied the effect of HIIT exercises on body composition and some metabolic

hormones in young men. The subjects participated in 16 weeks of intense periodic training (8 to 10 four-minute periods of running at the rate of 80% to 90% heart rate reserve ( $HR_{rest}$ )), with active rest intervals for two minutes of running at 40% to 50% heart rate reserve ( $HR_{rest}$ ), for three sessions a week and each session lasted for 50 to 60 minutes. The results showed that the fat percentage, waist circumference and waist-to-hip ratio were decreased significantly after exercise (17). Kordi et al. (2013) reported that a six week training protocol (three sessions per week, each session consisted of four to six reps running at maximum velocity in an area of 20 meters with 20 to 30 seconds of rest) reduces the fat percentage, BMI and waist-to-hip ratio in young women with sedentary life (18). Khodaei et al. (2014) reported that two weeks (five sessions a week) of HIIT training increased cardiovascular endurance in active women (19). The improvement of cardiorespiratory fitness could be due to improved transport mechanisms of oxygen delivery to the skeletal muscle by increasing the heart rate, stroke volume and cardiac output and also increasing blood volume, and injections deductions, reducing peripheral resistance, improving endothelium function in peripheral vascular relaxation and increasing peripheral blood flow and oxygen uptake and receiving by the cell and through increasing capillary and mitochondrial density (19). Increasing blood hemoglobin and enzymatic adaptation (increasing gene expression and enzyme activity involved in beta-oxidation, the Krebs cycle and the electron transport chain, such as citrate synthase) are among other mechanisms which are associated with increased cardiovascular endurance after HIIT trainings (19). Weight loss and fat percentage loss can also be due to the increasing the capacity of fat oxidation caused by increased gene expression and activity of lipolytic mitochondrial enzyme (18).

It was found in this study that HIIT training had no significant effect on E-selectin and P-selectin levels. In agreement with our result, Saetre et al. (2011) analyzed the effect of eight weeks (two sessions per week) walking workout on the levels of E-selectin and intercellular adhesion molecule 1 in 29 male and female patients with peripheral artery disease and reported the reduction of the variables (8). In a study, Bjornstad, et al. (2008) reported the positive effect of 20 weeks of resistance and endurance training on P-selectin levels in patients with chronic heart failure (6). Pizza et al. (2001) reported that four weeks of eccentric training did not make any significant changes in the level of E-selectin (20). Tanjes et al. (2007) suggested that four weeks of intense physical activity, not only reduce the fat percentage and weight, but also cause the reduction of E-selectin, vascular adhesion molecules 1 and intercellular adhesion molecule 1 in patients with diabetes

**Table 3.** Differences of Post-Test and Pre-Test Based on Paired T-Test Results in Each Group

Group	Tests	Weight, kg	BMI, kg/m <sup>2</sup>	CRF, s	E-Selectin, ng/mL	P-Selectin, ng/mL
Experimental	Pre-test	85.5 ± 16.6	33.4 ± 5.7	474.25 ± 57.8	25.07 ± 12	5.2 ± 2.1
	Post-test	83 ± 15.6	32.4 ± 5.2	366 ± 71.3	23.6 ± 11	5 ± 2.7
	P value	0.01 <sup>a</sup>	0.01 <sup>a</sup>	0.005 <sup>a</sup>	0.06 <sup>a</sup>	0.53
Control	Pre-test	76.2 ± 12.06	29.7 ± 4.2	473.7 ± 4.46	22.4 ± 4.2	4.1 ± 0.3
	Post-test	75.1 ± 11	29.3 ± 4	475.2 ± 3.8	23 ± 5.7	4.06 ± 0.86
	P value	0.44	0.46	0.17	0.6	0.77

Abbreviation: CRF (s), Cardiorespiratory Fitness.

<sup>a</sup>Unit: seconds of running activity.

**Table 4.** Comparison of the Results Between Groups Based on Independent T-Test Results

Variable	T-Score	df	P Value
Weight, kg	-0.96	14	0.35
BMI, kg/m <sup>2</sup>	-1.06	14	0.3
CR fitness	-4.05	14	0.001*
E-selectin, ng/mL	-1.6	14	0.12
P-selectin, ng/mL	-0.55	14	0.6

(21). Lim et al. (2015) reported that a combination of aerobic, resistance and traditional Korean dance exercises for 12 weeks (three days a week) decreased the E-selectin and vascular adhesion molecules 1 in old women (22). In a study by Jalaly et al. (2015), 12-week aerobic exercise reduced the levels of intercellular adhesion molecules 1 and E-selectin in patients with angina pectoris (23). In a study, Hamedinia and Haghghi (2007) investigated the effect of endurance and resistance exercise on circulating soluble adhesion molecules in obese men. The endurance exercises included running with the intensity of 75% to 85% of maximum heart rate (HR<sub>max</sub>) and resistance training included 11 stations of weight training and each session was done with four sets of 12 repetitions and the intensity of 50% to 60% of one maximum repetition. The results showed that both types of exercise decreased the E-selectin levels and intercellular adhesion molecules 1 (4).

The effective factors on E-selectin and P-selectin levels included age, body weight, BMI, body fat percentage, lipid levels and vascular stiffness (24, 25). As shown in this study, the levels of these substances after HIIT trainings were not changed in spite of decrease in BMI and weight. one limitation may be due to not measuring the changes in lipid levels; also E-selectin and P-selectin inside alpha granules of platelets and Weibel-Palade bodies of endothelial cells are stored and cytokines stimula-

tion and inflammatory mediators such as C-reactive protein, interleukin-1, interleukin-6, tumor necrosis factor-alpha, bacterial lipopolysaccharide, histamine, kinase keratin, complement components and free radicals can move them from the inside of platelets and endothelial cells to the endothelium surface and inside blood that causes the inflammation and platelet aggregation (24); and probably these mechanisms have not been fully effective in current study. Another possible mechanism is related to the lack E-selectin and P-selectin changes and the estrogen hormone fluctuations. Gene expression is suppressing the E-selectin and P-selectin and thereby is reducing the levels of these substances. since the subjects were young, and high levels of estrogen was absolutely available, which support the subjects in terms of E-selectin and P-selectin estrogen hormone is a protective hormone of heart and arteries, especially the coronary arteries and it is suppressing the gene expression of adhesion molecules, such as E-selectin and P-selectin (26, 27).

#### 4.1. Conclusion

In general, the results of current study showed that eight weeks of HIIT trainings had no significant effect on the levels of E-selectin and P-selectin of obese young women. For the reduction of E-selectin and P-selectin levels, the more intensified HIIT training was probably needed (each week more than three sessions or more than eight weeks). However, more researches needed to make more crucial decisions.

#### Footnote

**Conflicts of Interest:** We have nothing to declare.

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