Aerobic exercise training reduces inflammatory markers involved in atherosclerosis

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Abstract

Introduction: Adipose tissue and inflammatory factors play important role in occurrence of atherosclerosis in overweight women. The present study aimed to investigate the effect of 8 weeks of aerobic exercise training on risk factors involved in atherosclerosis in overweight women.

Materials and methods: For this, fourteen overweight women (mean ± standard deviation: body mass index 28.49 ± 3.28 k/m^2 and body fat 35.01 ± 3.68%) conducted aerobic exercise training for 8 weeks (at intensity correspond with 65 to 80 % reserved heart rate). Before and after aerobic exercise training, fasting blood samples were taken and anthropometrics characteristics were measured. The data were analyzed using Paired sample t-test ($P<0.05$).

Results: Aerobic exercise training significantly increased maximal oxygen consumption in overweight women ($P=0.001$). In contrast, aerobic exercise training result in significant reduction in intracellular adhesion molecule 1 levels ($P=0.013$) and C-reactive protein ($P=0.001$). In addition, anthropometric measurements of body fat percentage ($P=0.006$), waist to hip ratio ($P=0.01$), and body mass index ($P=0.001$) showed a significant reduction, too. However, no significant change observed in platelet ($P=0.127$), high density lipoprotein ($P=0.107$), low density lipoprotein ($P=0.095$) and cholesterol ($P=0.391$) levels.

Conclusion: Reduction in body fat following aerobic exercise training in overweight women is corresponding with a reduction in inflammatory markers involved in atherosclerosis.

Keywords: Aerobic exercise training, Intracellular Adhesion Molecule1, C-reactive protein, Body fat percent

Introduction

Epidemiological studies have raised inactivity and obesity as the most important risk factors involved in atherosclerosis (1-3). Increased fat tissue, particularly visceral fat, is associated with an increase in inflammatory biomarkers of intracellular adhesion molecule 1 (ICAM-1) and C-reactive protein (CRP) (4-7).

ICAM-1 plays a critical role in occurrence of atherosclerosis (4). ICAM-1 also known as CD54 is a protein with 90 kDa, a member of the immunoglobulin super family and a trans membrane protein that is encoded by ICAM1 gene in human (4, 6). ICAM-1 exacerbates inflammation by increasing the transmigration of leukocytes across vascular endothelial and recruitment of macrophages and granulocytes (8, 9). Upon CRP stimulation, concentration of ICAM-1

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greatly increases and expresses by the vascular endothelium, macrophages, and lymphocytes (5, 6, 10). Its increased expression result in more binding of low density lipoprotein (LDL) and platelet on endothelial cells, finally plaque is formed (10). In addition, calcium and cholesterol deposit on the plaque cause severe calcification and hardening of the arteries (4, 6, 10).

Overall, in this study as a model, the concentrations of ICAM-1, CRP, platelet, LDL and cholesterol were used as indirect biochemical markers for assessment of atherosclerosis (4, 6, 10). Among non-pharmacological strategies in improving cardiovascular diseases, exercise training has been considered effective (5, 11, 12). In this context, a reduction in serum ICAM-1 and CRP have been reported in diabetes mellitus type 2 and coronary artery patients following daily treadmill walking (70 to 85 % of maximum heart rate) (13) and cycle ergometer training (80 % of maximum heart rate) (7), respectively. However, in obese (14, 15) and overweight (16) individuals different effects of exercise training have been reported. In two studies last for one year, exercise training and nutrient modification result in a reduction in serum ICAM-1 and other inflammatory cytokines in obese individuals (14, 15). Also, a reduction in cholesterol and LDL in overweight women has been shown followed by 6 weeks of walking intervention (16). In contrast, Scheede-Bergdahl and co-workers have claimed 8 weeks of home-based exercise training program (rowing ergometer at 65 to 70 % maximal oxygen consumption) was not sufficient for improvement in inflammatory biomarker especially ICAM-1 in overweight subjects (17). Furthermore, resistance exercise training (18) and stationary cycle ergometer (19) for long duration had no significant effect on serum ICAM-1 and CRP concentrations of overweight (18) and obese (19) individual, respectively.

The annual rate of overweight and obesity growth has recently been going up worldwide and can be seen in both sexes and all races (4, 6, 10). In a national study of diseases in Iran, it is alleged that cardiovascular diseases are in third rank and atherosclerosis is higher in obese and overweight subjects (20). Furthermore, obese individuals have elevated levels of ICAM-1 compared to lean ones (21). Due to lack of cohesion information regarding the health effects of exercise training on overweight patients, the results of present study will give us new insights about real effect of exercise training on biomarkers involving in atherosclerosis diseases. Hence, in the present study the concentrations of ICAM-1, CRP, platelet, LDL and cholesterol was studied as indirect biochemical markers for assessment of atherosclerosis in overweight women.

Materials and methods

Fourteen sedentary overweight women (age 35 ± 6 years, mass 71.09±10.22 kg and height 157.79±5.36 cm) with regular menstrual cycle were selected after completing the healthy and Physical Activity Readiness Questionnaire (PAR-Q). Also, their physical fitness level was evaluated by Baecke's Physical Activity Questionnaire. All of them signed a written consent form while informing, the potential benefits, and the study associated-risks. According to Bielinski and colleague (2008) investigation, our exclusion criteria included any cardiovascular disease, atherosclerosis, hyperlipidemia, hypertension, diabetes, cancer and family background of these diseases (22). Also, we excluded any subject who smoke or use cholesterol-lowering medications, and non-steroidal anti-inflammatory drugs. The present study was approved by the Human Subjects Protection Committee of the University of Birjand (Iran).

Skinfold 3 site thickness was obtained with skinfold caliper (Yagami model,

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Japan) on the right side of the subject's body, using Jackson and Pollock equation (triceps, thigh and suprailiac) to estimate body density, while body fat percentage (BF %) is subsequently calculated using the Siri equation. Waist-hip ratio (WHR) is calculated as waist measurement (at the smallest circumference of the waist, just above the belly button) divided by hip measurement (its widest part of the buttocks). The subject's height and weight are measured by digital stadiometer Seca. Body mass index (BMI) is calculated as weight in kilograms divided by the square of height in meters.

Participants are appropriately trained under physician supervision. Participants run for 8 weeks (1 session per day, 4 days per week). Before and after each exercise session, subjects perform warm-up and cool-down exercises for 15 minutes. The training started with intensity correspond to 65 % heart rate reserve for 16 min. Running duration increased 2 min each week until reaches to 30 min at 8th weeks. Also, intensity increased 5 percent each two weeks until reaches 85 % heart rate reserve at 8th week. Karvonen formula was used to calculate heart rate reserve as: [(max HR − resting HR) × % intensity] + resting HR. Also, subjects maximal oxygen consumption (VO_2max) determined by Storer-Davis maximal test on ergo meter before and after training protocol.

To avoid facing with hormonal changes of menstrual cycle, sampling time coincides with the luteal phase because estrogen acts as an anti-inflammatory factor which reduces serum ICAM-1 (23). Blood samples were obtained after 12 hours fasting from the antecubital vein before and 48 hours after last resistance exercise training session (18). The samples were centrifuged (Eppendorf Centrifuge, Mini Spin R, Germany) for 10 min at 3000 ×g, at 4 °C. Serum was collected and stored immediately at −80 °C. Serum ICAM-1 (Gen-Probe Diaclone SAS, France) and CRP (Mississauga, Ontario, Canada) levels was measured using the commercially ELISA kit according to the manufacturer's instructions. The sensitivity of the ICAM-1 and CRP kits were less than 8.68 ng/ml and 10 ng/ml, respectively. High density lipoprotein (HDL), and total cholesterol (TC) levels were measured by enzymatic methods (Parsazmoon Co., Karaj, Iran). Finally, serum LDL levels were determined by Friedewald equation.

Statistical analysis

Data were analyzed by statistical package for social sciences (SPSS Inc., Chicago, USA) version 16.0 and expressed as mean ± standard deviation (SD). After determination of normal distribution of data by Shapiro-Wilk's test, they were statistically analyzed by Paired sample t-test. Significance level was set at $P < 0.05$.

Results

VO_2max, as performance characteristic, significantly increased following aerobic training (18.23±3.34 vs. 23.88±4.01 ml/kg/min for pre and post, respectively) (t_{13}=8.14, P=0.001) in overweight women (Figure 1).

Aerobic exercise training significantly reduced atherosclerosis biochemical markers of serum levels of ICAM-1 in post test (505.07±103.37ng/ml) compare to pre test (590.71±93.27 ng/ml) (t13=4.05, P=0.001) (Figure 2A). As depicted in Figure 1, serum levels of CRP reduced significantly following aerobic training (1368.5±839.65 vs. 945.43±734.66 ng/ml for pre to post test, respectively) (t_{13}=9.60, P=0.001) (Figure 2B).

In addition, aerobic exercise training significantly reduced anthropometric characteristics of body mass (71.09±10.22 vs. 68.04±10.07 kg for pre to post, respectively) (t_{13}=5.82, P=0.001) (Figure 3A), BMI (28.49±3.28 vs. 27.20±3.25 kg/m2 for pre to post, respectively) (t_{13}=5.30, P=0.001) (Figure 3B), BF%

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(35.01±3.68 vs. 33.87±3.06 for pre to post, respectively) (t_{13}=3.24, P=0.006) (Figure 3C) and WHR (0.81±0.03 vs. 0.79±0.03 for pre to post, respectively)(t_{13}=2.99, P=0.010)(Figure 3D).

In contrast, aerobic exercise training had no significant effect on serum lipids of LDL (95.50±16.72 vs. 107.43±20.42 mmol/l for pre to post, respectively) (t_{13}=1.802, P=0.095) (Figure 4A), HDL (48.64±7.69 vs. 44.28±8.39 mmol/l for pre to post, respectively) (t_{13}=1.73, P=0.107) (Figure 4B), and TC (176.50±25.16 vs. 182.86±34.92 mg/dl for pre to post, respectively) (t_{13}=0.88, P=0.391) (Figure 4C) in overweight women. Also, aerobic exercise training had no significant effect on platelet counts (22450±17027 vs. 23843±18143 count/mm3 for pre to post, respectively) (t_{13}=1.63, P=0.127) (Figure 4D) in overweight women.

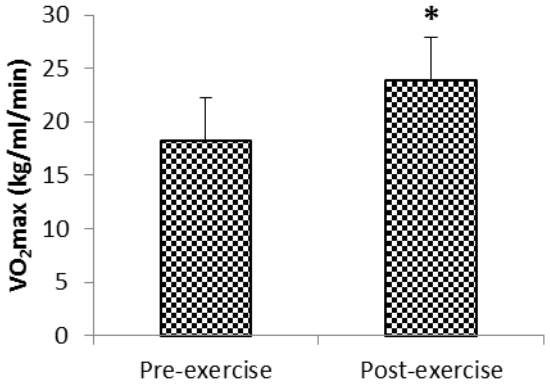


Figure 1. 8 weeks of aerobic exercise training significantly increased VO_2mas (P=0.001) in overweight women. Abbreviations: V_{O2}max; Maximal oxygen consumption. * indicate significant difference with Pre-exercise (P <0.05).

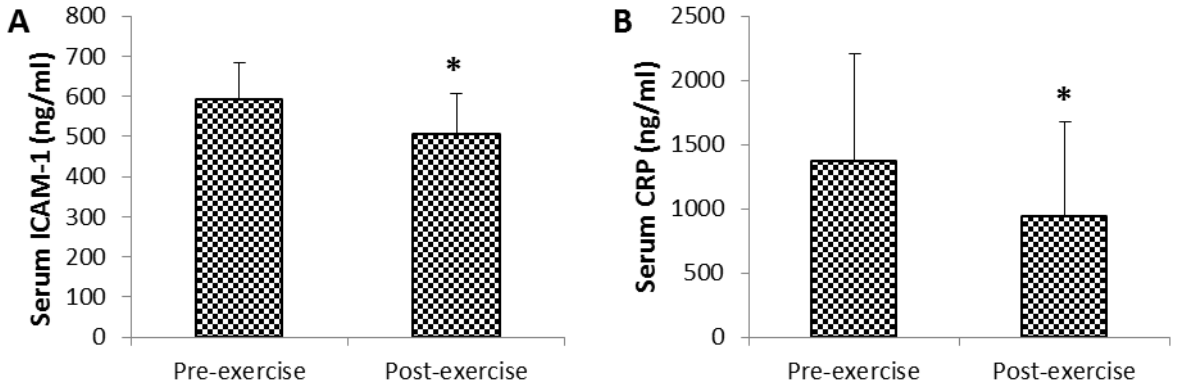


Figure 2. 8 weeks of aerobic exercise training significantly reduced serum ICAM-1 (P=0.001) (A) and CRP (P=0.001) (B) in overweight women. Abbreviations**:** ICAM-1; Intracellular Adhesion Molecule 1, CRP; C - reactive protein.

* indicate significant difference with Pre-exercise (P <0.05).

Discussion

Although increased LDL and decreased HDL parameters have been considered to determine the risk for cardiovascular disease, the reports show that people, who had been suffering from cardiovascular disease, have normal levels of LDL and HDL (6, 10). So, a lot of researches have been suggested that cardiovascular

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diseases have inflammatory background and systemic inflammation plays a pivotal role in its development (4, 5). The results of present study revealed that aerobic exercise training for 8 weeks has no significant effect on HDL, LDL, and cholesterol, while serum levels of ICAM-1 and CRP as two inflammatory characteristics reduced in overweight women.

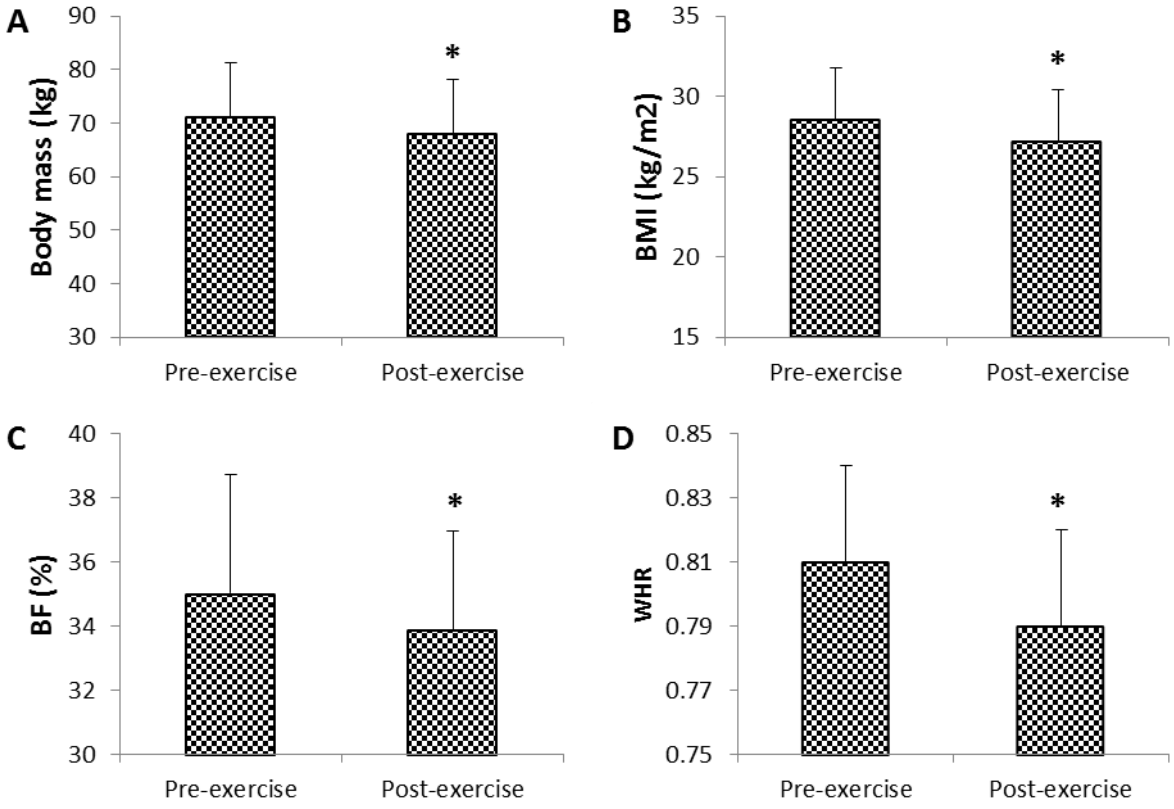


Figure 3. 8 weeks of aerobic exercise training significantly reduced serum body mass (P=0.001) (A), BMI (P=0.001) (B), BF % (P=0.006) (C), and WHR (P=0.010) (D) in overweight women. Abbreviations**:** BMI; Body mass index, BF %; body fat percentage, WHR; Waist-hip ratio.
* indicate significant difference with Pre-exercise (P <0.05).

Oxidation of LDL (LDL-ox) followed by deposition on endothelial cells, up-regulates ICAM-1 expression (16). Also, reduced flexibility and increased fragility of vessels by cholesterol can occur in presence of calcium (7). Our findings are inconsistent with other studies, such as consuming 3 to 6 weeks of raisins (16) and caloric restriction (13) along with exercise training, resulted in reduction of LDL and cholesterol concentrations in overweight (16), and diabetic patients (13), respectively. It has been demonstrated that raisins act as anti-atherosclerosis through interfere with cholesterol absorption (16).

Other aspects of our model outline about risk factors involved in atherosclerotic disease are platelets. Platelets Binding to ICAM-1 result in plaque formation and subsequently vascular occlusion (6, 10). Our results revealed that aerobic exercise training for eight weeks has no significant effect on platelets concentration. Therefore, no change in platelet components in assumed model considers a positive mechanism at least here. Effect of exercise training on endothelial function is mediated through stimulation of prostaglandin (PGL_2) (24). PGL_2 secrets from vessel wall or smooth muscle cell, in turn inhibits platelet aggregation and

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finally led to reduced expression of ICAM-1 (24). Due to study limitations, the activity of immune cells and platelet adhesion was not measured in present study, so it is recommended to be considered in future researches. There is now a substantial body of evidence suggesting that aerobic exercise training is a significant factor against occurrence of atherosclerosis through attenuation of inflammatory cytokines (7, 13, 16).

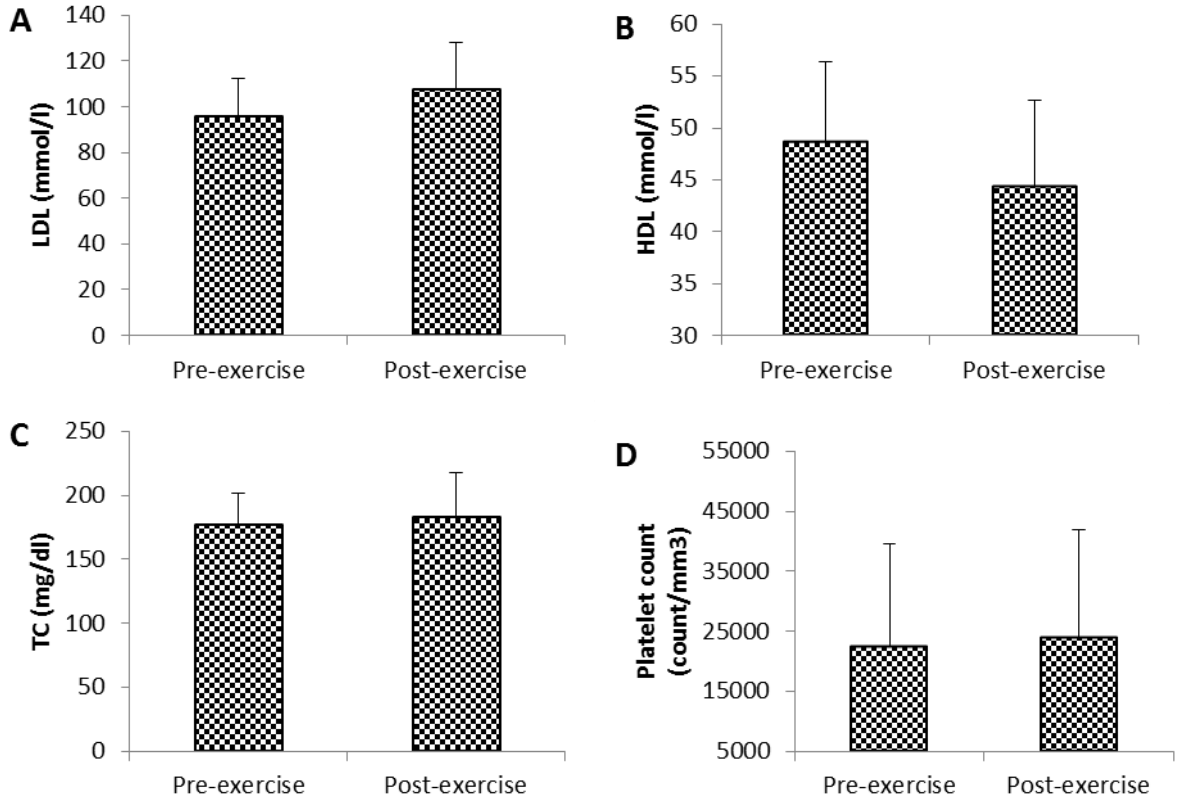


Figure 4. 8 weeks of aerobic exercise training had not significant effect on serum LDL (P=0.095) (A), HDL (P=0.107) (B), TC (P=0.391) (C), and platelet count (P=0.127) (D) in overweight women. Abbreviations: LDL; Low Density Lipoprotein, HDL; High Density Lipoprotein, TC; Total Cholesterol.

In contrast to other aspects of assumed model, showing no change happened; we showed a reduction in ICAM-1 and CRP in line with the decline in BMI and BF (%). Our findings are consistent with other studies that revealed a reduction in ICAM-1, CRP, and BMI following nutritional interventions and exercise training (7, 13). Also, in two studies a reduction in ICAM-1 attributed to artery remodeling after exercise training without weight loss in sedentary overweight adults (15, 19). In addition, nutritional interventions through reduction in dietary fat and increasing the dietary fiber amount, affected the adipose tissue which subsequently reduced the ICAM-1 expression (14). Especially, the visceral adipose tissue loss in obese women, lead to more significant improvement in inflammatory cytokines and ICAM-1 expression (14). In this context, a positive correlation between ICAM-1 with BMI and BF (%) in type 2 diabetes patients has been showed (25). Also, it is consistent with the study of Puglisi and co-workers (2008) which attributed a reduction in plasma ICAM-1 following 6-week of walking in overweight postmenopausal women (16). As our finding showed a decline in ICAM-1 in obese women, one study reported a reduction in ICAM-1 in obese men

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following daily treadmill walking (13). It should be noted that serum ICAM-1 level is not different between men and women (21). In contrast, 8 to 14 weeks of rowing on a home rowing ergometer (17) and stationary cycle training (19) had not any significant effect on concentration of ICAM-1 and CRP. This lack of change appears to be resulted from inability of mentioned exercises in influencing the body fat. Although it seems that artery remodeling induced by exercise training results from less shear stress and ICAM-1 expression (15), it seems that major determinant of changes in ICAM-1 and CRP levels in overweight and obese women to be body fat, particularly visceral fat. Adipose tissue by producing inflammatory cytokines such as tumor necrosis factor (TNF-α) and interleukin-1 (IL-1), may increase expression of ICAM-1 on endothelial cells (7, 13-15). Inflammatory cytokines enhance NF-kappaB activity that can regulate transcription of ICAM-1 (26). Moreover, NF-kappaB promotes the enhanced attachment of monocytes and macrophages to vessel walls through synthesis and releasing the pro-inflammatory cytokines (26). Importantly, TNF-α and IL-1 stimulate synthesis of CRP in hepatocyte, while CRP increases ICAM-1 expression (4, 5).

Furthermore, Olson and colleague reported no changes in ICAM-1 amount following one-year of moderate resistance training in overweight women attributed to increased adiponectin and reduced CRP levels (18). In fact, adiponectin (18) and estrogen (14, 16, 23) act as anti-atherosclerotic markers and it has been shown that exercise training improve adiponectin (18) and estrogen (14, 16) levels. In other words, exercise training by increasing estrogen (16, 23) and adiponectin levels and anti-inflammatory cytokine (16, 18) reduces the expression of ICAM-1 and CRP induced by inflammatory cytokines such as IL-1 and TNF-α which release from adipose tissue. Moreover, exercise training improves antioxidant capacity; subsequently reduce free radical production and LDL oxidation. In addition, decreased ICAM-1 expression associates with reduction in oxidation of LDL (16). Finally, it seems that part of the observed decline in levels of ICAM-1 in present study is due to reduced shear stress induced by angiogenesis and arterio genesis followed by exercise training (15, 19). Shear stress results in cleavage of mICAM-1 and its release to circulation (19).

Conclusion

In the present study, as a model, the concentrations of ICAM-1, CRP, platelet, LDL and cholesterol was used as indirect biochemical markers for assessment of atherosclerosis. Although, HDL, LDL, cholesterol and platelets as parts of assumed model did not change following 8 weeks of aerobic exercise training, other components of mentioned model i.e. inflammatory risk factors (ICAM-1 and CRP) had been reduced in overweight women. Furthermore, it seems that reduction in BMI and percent body fat may stimulate the reduction in inflammatory markers involved in atherosclerosis, following 8 weeks of aerobic exercise training in overweight women. Indeed, aerobic exercise training can be recommended for overweight women, because of reduction in both anthropometric characteristics and inflammatory markers.

Acknowledgments

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Conflict of Interests

Authors have no conflict of interests.

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