

A comparison of the effects of two weight loss protocols on plasma concentration of IL-6 in female sedentary college students

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Received 16 June 2010

Accepted 7 August 2010

Abstract

Interleukin-6 (IL-6) is excreting by adiposities, which increases by overweight and obesity. Obesity-related inflammatory marker is a risk factor for diseases. It has been reported that diet-induced weight loss lowers IL-6 levels. The effect of exercise training and other therapies on IL-6 is still unclear. We examined the effects of two weight loss protocols on plasma concentration of IL-6 in a sample of overweight and obese healthy sedentary female college students (N = 30; BF = 29.53±3.1%, and BMI = 29.04±2.22 kg/m²), who were randomly assigned into three experimental and one control groups. Experimental Group I experienced twelve-day researchers' proposed diet (D); Experimental Group II experienced twelve-day researchers' proposed diet along with twelve-day running training every other day mid consumption 250cc water beverage during per aerobic exercise period (AEDW), Experimental Group III experienced twelve-day researchers' proposed diet along with twelve-day running training every other day and the consumption of 250cc 12% glucose beverage during each aerobic exercise (AEDG). The control group (C) remained sedentary with a normal diet. Total body weight in all three experimental groups was reduced ($p < 0.001$). Also, IL-6 levels were significantly decreased, for both AEDW and AEDG groups ($f = 22.962$, $p = 0.022$) in comparison with the control group, but significant decrease in resting levels of IL-6 in AEDW group was more than AEDG ($p < 0.001$ vs. $p < 0.022$). There was no significant change in the D and control group ($f = 22.962$, $p = 0.781$). After doing the exercise and enforcing diet protocols, AEDW and AEDG groups lost weight and showed a decrease in their resting IL-6 plasma concentration. These protocols are optional for decreasing resting plasma IL-6 levels and prevent prevalence disease as cardiovascular disease.

Keywords: Interleukin-6, Aerobic exercise, weight loss, overweight.

Introduction

The 1998 clinical guidelines for the treatment of overweight and obesity issued by National Heart, Lung, and Blood Institute (NHLBI) divided adults into six categories on the basis of body mass index (BMI) (e.g., 25 to 29 as overweight; 30 to 34.9 as middle obese). The argument for health risks in the newly labeled overweight category was based more on morbidity than on mortality, which explains why the individuals with BMI in this range were labeled *overweight*, not *obese* [1]. Overweight and obesity cause or exacerbate a large number of health problems, both independently and in association with other diseases, and are among the most significant contributors to illness [2]. IL-6 belongs to the IL-6 family of cytokines, including IL-11,

leukemia inhibitory factor, ciliary neurotrophic factor, cardiotrophin-1, and cardiotrophin-like cytokine [3] and excreting by adiposities, which increases in overweight and obese people [4]. One possible explanation for elevated inflammatory markers in obesity is that adipose tissue secretes a number of pro-inflammatory cytokines, including interleukin-6 (IL-6). IL-6 is an adipocyte secretory product that may be involved in insulin resistance, which is a cytokine secreted by many cells, including adipocytes and adipose stromal cells. Adipose tissue has been proposed as a factor directly modulating pro-inflammatory and anti-inflammatory cytokine levels [2, 5, 6]. These factors contribute to the release of free fatty acids (FFAs) from abdominal adipocytes into the portal system [2].

Likely IL-6 inhibits the expression of lipoprotein lipase (LPL) in adipocytes, but IL-6 does not stimulate lipolysis [8,9]. Elevated levels of several

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proinflammatory cytokines, such as IL-6, have been found associated with proxy indicators of elevated body fat (body weight and body mass index) and with cardiovascular disease risk factors [4]. Moreover, several pro-inflammatory molecules such as IL-6 have been prospectively associated with thrombotic cardiovascular events [5]. IL-6 has been shown to play an independent role in the development of atherothrombosis and thus may represent a mechanistic link between obesity and the development of coronary heart disease and overall cardiovascular disease (CVD) elevated levels of inflammation also appear to directly mediate the expression of cell adhesion molecules, which have been linked to atheromaformation [10]. Esposito et al (2003) reported that positive correlations among body weight, BMI, WHR, and levels of IL-6, which suggests that the circulating levels of these cytokines may reflect, at least in part, production by adipose tissue [11].

Obesity is recognized as a major risk factor for cardiovascular diseases (CVD) including coronary heart disease. Individuals who are obese commonly demonstrate elevated levels of blood marker suggesting, chronic low-grade systemic inflammation. Altered plasma concentrations of inflammatory mediators may be pathogenic by inducing systemic endothelial dysfunction. Several studies have demonstrated that elevated markers of inflammation such as IL-6 [11, 12] are predictors of insulin resistance. It has been suggested that exercise produces a short term inflammatory response, whereas both cross-sectional comparisons and longitudinal exercise training studies demonstrate a long-term anti-inflammatory effect (25). Obesity has become a national epidemic with enormous public health implications, and recent studies have demonstrated a further 6% increase in the incidence of obesity [body mass index (BMI) .30. kg/m²] over a 7-year period [9]. Overweight and obesity is highest in middle age and older adults and is thought to contribute to the increased risk of coronary artery disease, cerebrovascular disease, and atherothrombotic events in this segment of the population [15]. It has been consistently demonstrated that the plasma concentration of IL-6 increases during muscular exercise as well as the following exercise; the basal plasma IL-6 concentration may increase up to 100-fold (acute response), but fewer dramatic increases

are more frequent. Today, it is clear that the contracting skeletal muscle is the main source of the IL-6 in the circulation in response to direct exercise. Whereas supplementation with carbohydrates during exercise inhibits the exercise-induced increase of IL-6 in plasma, IL-6 mRNA expression within the contracting muscle is unaffected [3].

The aim of the present study was to compare the influence of two weight loss protocols on plasma concentration of IL-6. Regardless of a few research which has investigated the effect of AED on the resting levels of plasma IL-6 concentration in overweight and obese individuals, most of the research has examined the effect of aerobic exercise on inflammatory markers of Cardiovascular Diseases such as C - reactive protein in patients or it has tested the effects of exercise on muscle and plasma concentration of IL-6. Therefore, assuming that AED would reduce the levels of IL-6, we tested the effects of AEDW, AEDG and D protocols on rest levels of plasma IL-6 in overweight and obese health sedentary female college students.

Methods

Participants

Thirty overweight and obese healthy sedentary female college students were selected and randomly divided into a control group or one of the three experimental groups: (BF% = 29.53±3.18 and BMI = 29.04±2.22 kg/m²).

Diet Protocol for Weight Reduction

We used a twelve-day diet weight reduction protocol, including researchers' proposed diet (Table 1). The weight loss method was extracted from Rashid Lamir et al.'s weight loss method [16].

Aerobic Exercise Protocol

AEDW and AEDG groups experienced a twelve-day aerobic exercise every other day along with the diet recommended by the researchers (the training intensity was 60-70 % HR max during 50 minutes).

Laboratory Measurements

Blood samples (5cc from-brachial vein) were collected at 8am (the participants were fasting for 12-13 hour prior to the test), 24 hours before the first and after the last day of the protocol. Blood

Table 1: reduction in food intake for overweight and obesity to lose weight

	1 st day	2 nd day	3 rd day	4 th day	5 th day	6 th day	7 th day	8 th day	9 th day	10 th day	11 th day	12 th day
Reduction in lunch	10%	10%	10%	Eating as usual	20%	20%	20%	10%	30%	30%	30%	20%
Reduction in dinner	10%	10%	10%	Eating as usual	20%	20%	20%	10%	30%	30%	30%	20%

samples for IL-6 measurement were drawn into pre-cooled glass tubes containing EDTA. The plasma was stored at -80°C until analyses were performed. After plasma separation, IL-6 levels were measured using ELISA method with a special kit. Body fat percentage of participants was measured using 3-points skin fold method.

Statistical Analysis

The data was analyzed by K-S, ANOVA and Tukey *post hoc*. All calculations were conducted using SPSS/PC, version 16.

Results

Baseline physical and metabolic characteristics of all the participants are shown in Table 1.

Body mass index (BMI) was ranged from 25.03

– 33.04 kg/m^2 . From pre- to post-test measurements, body weight was reduced for AEDW, AEDG and D groups (F (df1, df2) = 27.042; $p = 0.000$), in comparison with the control group. Body fat percentage (BF %) in AEDW ($F = 61.82$; $p = 0.000$) and AEDG ($F = 61.82$; $p = 0.012$) groups were decreased in comparison with the control group. However, there was no significant change in BF % in D group ($F = 61.82$, $p = 0.114$) in comparison with the control group. Resting Plasma IL-6 levels of overweight and obese female population were also reduced for both AEDW ($F = 22.96$, $p = 0.001$) and AEDG ($F = 22.96$, $p = 0.022$) groups, in compare with the control group. There was no significant change in D group ($F = 22.96$, $p = 0.781$) in comparison with the control group (Table 2 and Table 3).

Table 1: Baseline Physical and Metabolic Characteristics in 30 overweight and obese healthy sedentary female of college students

	Minimum	Maximum	Mean	SD
Height	150.00	170.00	1.61	4.79
Weight (kg)	66.10	86.70	75.60	5.57
BMI (kg/m^2)	25.03	33.04	29.04	2.22
Body fat %	24.80	38.30	29.53	3.18
Plasma concentration of IL-6 (pg/ml)	0.06	0.23	0.15	0.05

Table 2: ANOVA for all participants (N = 30)

Factor		F-Value	p-Value
Plasma concentration of IL-6 (pg/ml)	Between Groups	22.962	0.001
Weight	Between Groups	27.042	0.001
BMI	Between Groups	25.709	0.001
Body fat %	Between Groups	61.821	0.001

Table 3: Tukey *post hoc* test for all participants (N = 30)

Factor	Groups			p-Value
	CONTROL	AED		
Plasma IL-6 levels (pg/ml)	CONTROL	AED	Along with water	0.001
			Along with glucose soluble	0.022
		D	0.781	
Body weight	CONTROL	AED	Along with water	0.001
			Along with glucose soluble	0.001
		D	0.001	
Body fat %	CONTROL	AED	Along with water	0.001
			Along with glucose soluble	0.012
		D	0.114	
BMI	CONTROL	AED	Along with water	0.001
			Along with glucose soluble	0.001
		D	0.001	

Discussion

This was the first study to examine the effect of weight loss protocol on plasma concentration of interleukin-6 in overweight and obese healthy sedentary female college students. The main finding of the research was an obvious reduction in plasma concentration of IL-6 in the experimental

groups followed by the completion of their weight loss protocol (AED). After the implementation of the programs, body weight was significantly reduced for AEDW and AEDG and D groups in comparison with the control group. Moreover, the plasma IL-6 levels were also reduced in AEDW and AEDG groups (Figure 1).

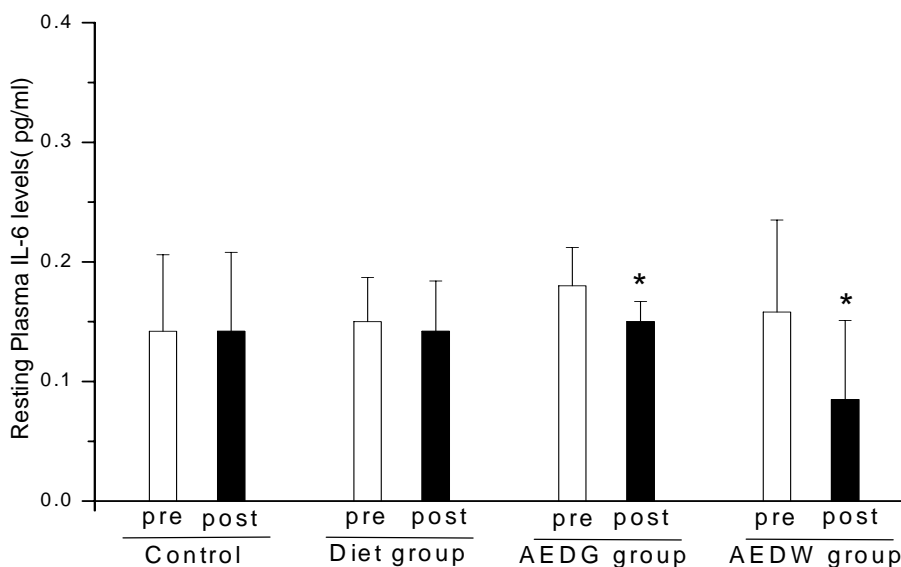


Figure 1: Resting Plasma IL-6 levels of the four groups before and after the completion of the intervention protocols.

These findings support the study hypothesis and are compatible with previous findings who demonstrated that a moderate weight loss improves insulin sensitivity and decreases serum concentrations of IL-6 due to weight loss [17]. Furthermore, Bastard et al. reported that IL-6 was reduced by weight loss after a very low-calorie diet that had been imposed on eight lean and 21 obese women for three weeks [18]. However, our finding about the D group did not show a significant change in resting plasma concentration of IL-6; this might be due to a short period of diet in our protocol, and the fact that IL-6 and BF% were decreased, for both AEDW and AEDG groups, but the significant levels of reduced IL-6 in AEDW were more than the levels in the AEDG group (0.073 vs. 0.03 pg/ml \pm for IL-6). There was no significant change in the mentioned variables in the mere diet group. This is could be because the amount of reduction of IL-6 and BF% in the AEDG group, who consumed glucose beverage during aerobic exercise, has not been enough. There are other studies that could show consuming carbohydrates during exercise inhibit training-induced increases in plasma concentration of IL-6

[5, 12, 15, 16, 18]. However, there are other studies that could not show changes in IL-6 plasma concentrations after regular physical exercise interventions either [13,14]. We found a change of plasma concentration of IL-6 after completing the AED protocol, due to the glucose beverage consuming that prevents the aerobic exercise-induced increase in plasma concentration of IL-6. Philip et al. (2001) and Koichi et al. (2004) reported that suppression in exercise-induced IL-6 can inhibit lipolysis in adiposities during exercise. Similarly, IL-6 inhibits the expression of lipoprotein lipase (LPL) and stimulates lipolysis in adipocytes. There are supporting data in favor of our results, indicating that the decrease of BF% in the AEDG group (who consumed soluble glucose during aerobic exercise) can suppress the aerobic exercise-induced increase in plasma concentration of IL-6. Consequently, the release of free fatty acids (FFAs) from abdominal adipocytes into the portal system [2] reduced, which, in turn, stimulates lipolysis in adipocytes [8,9] during and after aerobic exercise. A close association between adipose tissue synthesis and secretion of inflammatory cytokines has previously been

demonstrated [14]. Previous investigations examined the ability of exercise to alter inflammatory markers while they have led to confusing results. These disparities may be related to the acute and chronic phases of the inflammatory response to exercise training [8, 9].

According to other research, overweight and obesity accelerated diseases such as atherosclerosis and myocardial infarction; excess adiposity has been linked with diseases (11); moreover, inflammatory mediators such as IL-6 have been linked to arterial thrombogenesis [23]. Based on our findings, a decrease of plasma concentrations of IL-6 at rest prevented prevalence diseases such as cardiovascular disease risk in overweight and obese individuals who are susceptible to these diseases. It has been demonstrated that exercise training with weight reduction had an effect on obesity-related inflammatory markers and conventional cardiovascular risk factors [14]. Gary et al. (2005) suggested the decrease of IL-6 with moderate aerobic exercise [15]. Overweight and obesity have been associated with increased cardiovascular morbidity and mortality. It has been suggested that increased inflammatory burden may contribute to the heightened risk of atherothrombosis. Fasting plasma IL-6 concentrations are positively related to adiposity [1, 15] our results are in agreement with other studies.

In summary, our findings indicate that aerobic training in combination with diet reduced the resting levels of plasma concentration IL-6 of overweight and obese healthy sedentary individuals. The present study supports the idea that resulting weight loss may help prevent co-morbid prevalent diseases among healthy overweight and obese individuals. Aerobic exercise and diet in addition to weight loss and reduced body fat percentage can significantly reduce plasma concentration of IL-6 in overweight and obese individuals. AED prevents the morbidity risks associated with overweight and obesity by reducing weight and adipocytes and also alleviating inflammatory markers such as IL-6. We recommend that overweight and obese individuals use our AEDW method to decrease BF%, weight and IL-6 more efficiently.

References:

- 1-Strawbridge WJ, Wallhagen MI, Shema SJ (2000). New NHLBI clinical guidelines for obesity and overweight: will they promote health? *Am J Public Health* 90: 340-343.
- 2-Kopelman P (2007). Health risks associated with overweight and obesity. *Journal compilation* © The International Association for the Study of Obesity. *Obesity Reviews* 8: 13–17.
- 3-Bente K, Pedersen A, Febbraio (2007). Muscle as an Endocrine Organ: Focus on Muscle-Derived Interleukin-6. *Physiol Rev* 88: 1379–1406.
- 4-Vozarova B, Weyer C, Hanson K, Tataranni PA, Bogardus C, Pratley RE (2001). Circulating interleukin-6 in relation to adiposity, insulin action, and insulin secretion. *Obes Res* 9:414–417.
- 5-Esposito K, Pontillo A, Di Palo C, Giugliano G, Masella M, Marfella R, et al (2003). Effect of weight loss and lifestyle changes on vascular inflammatory markers in obese women: a randomized trial. *JAMA* 289: 1799–1804.
- 6-Ziccardi P, Nappo F, Giugliano G, et al (2002). Reduction of inflammatory cytokine concentrations and improvement of endothelial functions in obese women after weight loss over one year. *Circulation* 105: 804–809.
- 7-Pischon N, Heng N, Bernimoulin JP, Kleber BM, Willich SN, Pischon T (2007). Obesity, inflammation, and periodontal disease. *J Dent Res* 86: 400–409.
- 8-Okita K, Nishijima H, Murakami T, et al (2004). Can exercise training with weight loss lower serum C-reactive protein levels? *Arterioscler Thromb Vasc Biol* 24:1868-1873.
- 9-Kern PA, Ranganathan S, Li C, et al (2001). Adipose tissue tumor necrosis factor and interleukin-6 expression in human obesity and insulin resistance. *Am J Physiol Endocrinol Metab* 280: 745–751.
- 10-Olson TP, Dengel DR, Leon AS, Schmitz KH (2007). Changes in inflammatory biomarkers following one-year of moderate resistance training in overweight women. *International Journal of Obesity* 31: 996–1003.
- 11-Cornelissen VA, Fagard RH (2005). Effects of endurance training on blood pressure, blood pressure-regulating mechanism, and cardiovascular risk factors. *Hypertension* 46:667-675.
- 12-Fernandez-Real JM, Broch M, Vendrell J, Gutierrez C, Casamitjana R, Pugeat M, Richart C & Ricart W (2000). Interleukin-6 gene polymorphism and insulin sensitivity. *Diabetes* 49: 534–537.
- 13-Nicklas BJ, Ambrosius W, Messier SP, Miller GD, Penninx BWJH, Loeser RF, Palla S, Bleecker E & Pahor M (2004). Diet-induced weight loss, exercise, and chronic inflammation in older, obese adults: a randomized controlled clinical trial. *American Journal of Clinical Nutrition* 79: 544–551.
- 14-Ryan AS, Nicklas BJ, Berman DM, Elahi D (2003). Adiponectin levels do not change with moderate dietary induced weight loss and exercise in obese postmenopausal women. *International Journal of Obesity Related Metabolic Disorders* 27: 1066–1071.
- 15-Gary P, Van Guilder, Greta L, Derek T, et al (2005). Endothelial t-PA release is impaired in overweight and obese adults but can be improved with regular aerobic exercise. *Am J Physiol Endocrinol Metab* 289: 807–813.
- 16-Amir Rashid Lamir, Mahmood Goudarzi, Ali Asghar Ravasi, (2009). The effect of gradual and acute weight

- loss on strength and endurance of well trained wrestlers. *Word journal of sport sciences* 2 (4): 236-240.
- 17-Magdalena O, Barbara ZM, Piotr K, Joanna, et al (2006). The effect of weight loss on serum concentration of interleukine-6 (IL-6) and insulin resistance. *Endokrynologia Polska/Polish Journal of Endocrinology Tom Volume (57)*: 190-196.
- 18-Bastard JP, Jardel C, Bruckert E, et al (2000). Elevated levels of interleukin-6 are reduced in serum and subcutaneous adipose tissue of obese women after weight loss. *J Clin Endocrinol Metab* 85: 3338-3342.
- 19-Henson DA, Nieman DC, Nehlsen-Cannarella SL, Fagoaga OR, et al (2000). Influence of carbohydrate on cytokine and phagocytic responses to 2 h of rowing. *Med Sci Sports Exerc* 32: 1384-1389.
- 20-Lancaster GI, Jentjens RL, Moseley L, Jeukendrup AE, Gleeson M (2003). Effect of pre-exercise carbohydrate ingestion on plasma cytokine, stress hormone, neutrophil degranulation responses to continuous, high-intensity exercise. *Int J Sport Nutr Exerc Metab* 13: 436-453.
- 21-Li TL, Wu CL, Gleeson M, Williams C (2004). The effects of preexercise high carbohydrate meals with different glycemic indices on blood leukocyte redistribution, IL-6, hormonal responses during a subsequent prolonged exercise. *Int J Sport Nutr Exerc Metab* 14: 647-656.
- 22-Ehlsen-Canarella SL, Fagoaga OR, Nieman DC (1997). Carbohydrate and the cytokine response to 2.5 h of running. *J Appl Physiol* 82: 1662-1667.
- 23-Willerson JT and Ridker PM (2004). Inflammation as a cardiovascular risk factor. *Circulation* 109: II-2-10II.