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The effect of strength training on anti-inflammatory cytokines, cortisol and testosterone in overweight men

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ABSTRACT

Exercise training-induced increase in anti-inflammatory cytokines is one of methods proposed to reducing chronic inflammation. The purpose of the present study was to investigate the effects of 10 weeks strength training on plasma anti-inflammatory cytokines and their relationships with testosterone and cortisol hormone and body composition in youngs men. Nineteen sedentary men (age, 20-30 yr), volunteers to participant in the study, that randomly divided to two groups of strength training (n=10) and control (n=9) group. Blood sample were collected before and after 10 weeks from intervention, and the concentrations of plasma IL-4, IL-10 and testosterone were measured. Ten weeks strength training programme consists of 7 exercise in two sets with 8 repetitions at 70% of 1-RM in each exercise, and this is then as progressive changed to three sets with 8 repetitions at 80% of 1-RM in weeks 10. Dependent and independent t test, indicated that strength training increased upper and lower body strength, fat free mass, testosterone hormone and decreased cortisol hormone and body fat percent (P<0.05), but concentrations of plasma IL-4 and IL-10 not changed (P>0.05). In addition, Pearson's correlation indicated that no relationships were between changes in anti-inflammatory cytokines concentrations and changes in testosterone cortisol hormone and body composition (P>0.05). These results indicated that despite significant improvement of strength and body composition, 10 weeks strength training did not effect on anti-inflammatory cytokines in young men.

Key words: Inflammation, Cytokine, IL-4 and IL-10.

INTRODUCTION

Sedentary lifestyle increases the risk of cardiovascular diseases, diabetes and other diseases related to plasma concentration of inflammatory cytokines. Increase in the level of inflammatory mediators is associated with wide variety of chronic diseases [1]. From the historical point of view, inflammation is the natural response of body to a severe infection episode; however, chronic inflammation is a symptom of chronic infection [2]. During the last decade, inflammatory mechanisms have been identified to be of key importance in pathologic process of several health disorders including ischemic cardiovascular diseases [1, 2, 3], rectum cancer [4], heart attack [1, 2 , 5], type 2 diabetes [1, 2 , 6], chronic obstructive pulmonary disease [2, 7], and Alzheimer [1, 8]. Considering the widespread and harmful effects, finding the behavioral interventions to decrease the inflammation such as exercise training is of extreme importance. Therefore, it is thought that inflammatory pathway are potential therapeutic target for interventions to reduce disease and disability [9].

Although a few pharmacological interventions, such as statin, and angiotensin converting enzyme inhibitor, use decrease inflammation, as evidence by reducing the concentration of CRP, no current medical agents with anti-inflammatory effects is known to be used for treatment persistent inflammation in the non-elderly [9, 10]. Yet, to date, there is little definitive evidence for therapies that can effectively treat individuals with elevated markers of inflammation that are within the clinically normal range. On the other hand, lifestyle behavioural interventions, including changes in food/dietary intake and physical activity, may have clinically significant benefits for improving inflammation over the long-term [1, 9]. Similar intervention studies indicated the relationship between physical activities and the level of inflammatory markers especially in case of chronic diseases with rising inflammation condition [9, 11]. Specifically, lower inflammatory biomarker concentrations are observed in individuals who report performing more frequent and intense physical activity [9].

Altered levels of cytokines are not only seen in inflammatory disease; acute exercise has an effect on cytokine responses and inflammation in healthy individuals. Cytokine production can be affected by physiological factors present in exercise such as stress hormones, acidosis, oxidative stress, and heat among others [12]. In addition, cytokine response may vary by the type of exercise, intensity, duration, recovery between exercise bouts and training status [11, 12]. The effects of regular or chronic exercise on basal levels of inflammatory markers have been used to recommend exercise as an anti-inflammatory therapy [11, 13]. Compared to acute bouts of exercise, chronic exercise effects on inflammatory markers have been less investigated [12, 14, 15].

Cytokine responses to a one-session resistance exercise differ with the response to long-term resistance training. In case of severe responses, metabolic needs and muscular damages play a great role [12]. On the other hand, long-term resistance training change the mass of the body, metabolism and tissues' function [12, 15]. For instance, on bout of acute resistance exercise prompts the production of reactive oxygen species (ROS) [12]. Nonetheless, long-term resistance training increase the antioxidant capacity of the cell [9].

Regular exercise training ensures their preservative effects through inducing the expression of endogenous antioxidants and anti-inflammatory cytokines. Studies have revealed that exercise training not only reduce levels of pro-inflammatory cytokines of IL-1β, TNF-α, IL-6 and CRP, but also increases the concentration of anti-inflammatory cytokines of IL-1ra, IL-4, IL-10 and transforming growth factor β (TGF-β) [23, 30]. IL-1ra, IL-4, IL-10 and TGF-β are not only anti-inflammatory in nature but also suppress the production of pro-inflammatory cytokines IL-1, IL-6 and TNF-α [23, 30].

It is also revealed that resistance training may at least improved insulin resistance in two ways: first, by decreasing low grade systemic inflammation and second by improving glucose uptake by muscular cells [12]. These effects may in part be the result of improved body composition (increase in muscle mass), quality of the muscle mass and metabolic adaptations [9]. Although the effects of aerobic training on inflammatory markers have been attentioned more, a handful of resistance training studies have been conducted, with results largely negative. In a recent study, Abd El-Kader reported a decrease in the levels of serum IL-6 and TNF-α after 12 week resistance training program [16]. However, Ferreira et al. (2010) and Levinger et al. (2009) failed to found any change in the concentration of serum IL-6, IL-1β and TNF-α after a 10 week resistance training [17, 18]. Moreover, Brochu et al. (2009) demonstrated that resistance training does not improve inflammatory markers; but if weight loss occurs at the same time, significant improvement is found in inflammatory and metabolic parameters [19].

Prior studies have demonstrated that acute strength exercise transiently elevateds circulating concentration of anabolic and catabolic hormones (i.e. testosterone and cortisol), and cytokines includes IL-10, IL-1ra, IL-6 and IL-1β [21]. With the controversy of the role of testosterone and cortisol in circulation, one of the primary targets might be the modulating activities of immune cells in the circulation [20]. It is now accepted that, there is a significant inverse relationship between testosterone and IL-6 soluble receptors [21] i.e. the lower of the level of testosterone is associated with the more pro-inflammatory state. In vitro evidence demonstrates that testosterone may suppress expression of pro-inflammatory cytokines of IL-1, TNF-α and IL-6 but potentiate expression of IL-10 anti-inflammatory cytokine [21, 22]. Similarly, the age-induced decrease in the level of steroid hormones are considered as an important reason for the probable increase in pro-inflammatory markers [20, 21]. Yet, the interaction between endocrine and cytokine responses in trained and untrained individuals has not been elucidated [20, 23].

Most of the studies on strength exercise interventions on inflammatory markers were conducted on middle-aged and elderly patients thus generalization of their results to younger individuals could be somehow dubious. Therefore, given the potential advantages strength training have on health and in an attempt to identify the effects strength training on anti-inflammatory cytokines, the present study investigates the changes in the plasma concentration of IL-4 and IL10 and their relationship with cortisol and testosterone response and body composition changes in overweight young men after 10 weeks of strength training.

MATERIALS AND METHODS

Nineteen sedentary overweight (BMI>25 kg/m^2) men who had no regular exercise for at least 12 months were recruited into this study. Subjects were randomly assigned to one of the two groups: strength training group (n=10) or control group (n=10). All subjects were asked to complete a personal health and medical history questionnaire, which served as a screening tool. All subjects were non-smokers and had no history of any kind of medical condition that would prevent them from participating in the exercise intervention. The University's ethics committee approved the experimental procedures and study protocols, which were fully explained to all subjects. A written consent form was signed by each subject after having read and understood the details of the experiments.

Experimental design

Following familiarisation, subjects were asked to report to the laboratory for an additional test session designed to determine one-repetition maximum (1-RM) for seven exercises involving the upper and lower body. A detailed description of the 1-RM testing procedure can be found elsewhere (24).

Anthropometric measurements

BMI was calculated by dividing weight (kg) to the square height (m). Waist was measured between the lowest rib and the iliac crest. The hips were measured in their widest part in the pelvic area [25]. The waist to hip ratio was calculated through dividing waist to the hips. In order to calculate the percentage of body fat, the thickness of the subcutaneous fat in the three point three heads, stomach and upper pelvic of the subjects was measured using a calliper and then was estimated via Jackson and Pollack three point equation [25].

Blood sampling and analysis

Blood samples were obtained from all subjects at 0800 h after an overnight fast before and after 10 weeks from intervention. Post-training blood samples from subjects in the training groups were obtained 3-4 days after their last exercise session. On the days before the blood samples were taken, subjects were asked to consume a weight-maintenance diet for 3 days and to avoid strenuous exercise for 4 days.

For cytokines and hormones measurement, 5-ml blood was drawn into a glass tube and were centrifuged at the room temperature, separated and frozen at -80c and stored until subsequent analysis. IL-4, IL-10 (ELISA kits, Bender MEd systems, Austria), testosterone and cortisol (ELISA kits, IBL, Germany) were analyzed by commercially available enzyme-linked immunosorbent assay. The intra- and inter-assay coefficients of variation for cytokines were <10%.

Training program

The strength training program utilized in the present study was similar to that reported previously [27]. In brief, strength training was performed 3 days per week for 10 weeks, with 48–72 h of recovery between training sessions. The training was consisted of 7 exercise (chest press, shoulder press, lat pull-down, seated row, leg press, leg curl and lunge) in two sets with 8 repetitions at 70% of 1-RM in each exercise, and this is then as progressive changed to three sets with 8 repetitions at 75% of 1-RM in weeks 10 (table 1). Recovery between sets and exercises was standardized at 120 s, and an increase in resistance was warranted if two extra repetitions could be performed in the last set on two consecutive occasions, which promoted subjects to train proximally to ''momentary muscle failure'' by exercise completion [27]. Each training session for the resistance group commenced with a 5-min dynamic stretching warm-up routine, followed by the main session, and concluded with 5 min of stretching exercises. The control group did not do any physical activity.

Table 1. Per iodized strength exercise training program overview

	Weeks 1-2	Weeks 3-4	Weeks 5-6	Weeks 7-8	Weeks 9-10
Chest press	2 S × 10 R	2 S × 10 R	3 S × 8 R	3 S × 8 R	3 S × 8 R
Shoulder press	2 S × 10 R	2 S × 10 R	3 S × 8 R	3 S × 8 R	3 S × 8 R
Lat pull-down	2 S × 10 R	2 S × 10 R	3 S × 8 R	3 S × 8 R	3 S × 8 R
Seated row		2 S × 10 R	2 S × 8 R	2 S × 8 R	2 S × 8 R
Leg press	2 S × 10 R	2 S × 10 R	3 S × 8 R	3 S × 8 R	3 S × 8 R
Leg curl	2 S × 10 R	2 S × 10 R	3 S × 8 R	3 S × 8 R	3 S × 8 R
Lunge	-	-	-	2 S [a] × 8 R	4 S [a] × 8 R
Session intensity [b]	70	70	75	75	75
Session volume [c]	100	120	136	152	168
Session duration [d]	30	35	40	45	50

[a] Each set of lunge is for one leg, that is, 2 S = one set for each the right and left leg, 4 S = two sets for each the left and right leg. [b] Percentage of 10RM. [c] Total lifts performed. [d] Does not include dynamic warm-up and warm-down stretches.

Statistical analysis

Descriptive statistics were computed and distributions of all variables were assessed for normality. Since the data distribution was normal, t-test and Pearson correlation test was used in order to analyze the data. The mean values of variables obtained before and after 10 weeks in both groups were compared using the dependent ''t'' test. An independent ''t'' test was used for the comparison between the two groups. The relationships between variables at baseline and in response to training were determined using Pearson's correlation test. The level of significance in all statistical analyses was set at $P<0.05$.

RESULTS

Physiological characteristics of the participants in pre and post-test are presented in table 2. Before the intervention, there was no significant difference in the BMI, body fat percentage, WHR, 1RM of the bench and leg press and the plasma concentration of IL-4, IL-10, cortisol and testosterone ($P<0.05$).

Dependent and independent t tests revealed that strength training significantly increased the 1RM of the bench and leg press in the post-test ($P=0.00$) compared to the control group. Furthermore, strength training decrease the body fat percentage in the post-test significantly ($P<0.007$). However, BMI and WHR did not demonstrate a significant change during the study period ($P<0.05$) (table 2).

As showed in table 3, plasma concentration of cortisol significantly decreased and plasma concentration of testosterone significantly increased in strength training group ($P<0.05$), while no significant change in the control group was found. In addition, strength training caused non-significantly increase of IL-4 and IL-10 after 10 weeks ($P>0.05$) (table 3).

Table 2. Physiological characteristics (mean ± SD) of the groups in pre and post-test

Variable		Control group	Strength group
Body mass (kg)	Pre	83.4 ± 5.2	84.1 ± 5.1
	Post	84 ± 4.9	86.2 ± 5.6
BMI ($kg.m^{-2}$)	Pre	27.3 ± 1.6	29 ± 1.8
	Post	27.6 ± 1.5	28.7 ± 2
WHR	Pre	91.1 ± 2.1	91.4 ± 1.9
	Post	93.4 ± 2.9	89.6 ± 2.7
Body fat percent (%)	Pre	21.1 ± 2.1	22.7 ± 2.7
	Post	22.4 ± 1.6	19.8 ± 2.1*
Chest press (kg)	Pre	73.4 ± 4	74.7 ± 4.3
	Post	74 ± 5.7	95 ± 10.6*ǂ
Leg press (kg)	Pre	149.1 ± 10.4	142.8 ± 11
	Post	150.8 ± 11.2	170.6 ± 9.5*ǂ

**: Indicates significant difference between pre- and post-training values ($P<0.05$). ǂ: Indicates significant difference between control and strength group values ($P<0.05$).*

Table 3. Plasma hormones and cytokines concentrations (mean ± SD) of the groups in pre and post-test

Variable		Control group	Strength group
Testosterone ($ng.ml^{-1}$)	Pre	4.7 ± 1.9	5.1 ± 0.9
	Post	4.5 ± 2.3	8.8 ± 2.2*
Cortisole ($ng.ml^{-1}$)	Pre	151.3 ± 72.1	146.1 ± 67.4
	Post	155.1 ± 68.9	101.4 ± 34.6*
IL-4 ($pg.ml^{-1}$)	Pre	0.84 ± 0.21	0.79 ± 0.19
	Post	0.86 ± 5.7	0.87 ± 0.2
IL-10 ($pg.ml^{-1}$)	Pre	3.4 ± 0.8	3.1 ± 0.87
	Post	3.3 ± 0.9	3.5 ± 1.1

**: Indicates significant difference between pre- and post-training and control and strength group values ($P<0.05$).*
Pearson correlation coefficient demonstrated that there was no significant difference between the plasma concentration of cortisol with IL-4 ($P= 0.3$ and $R= 0.27$) and IL-10 ($P= 0.13$ and $R= 0.37$) and the plasma concentration of testosterone with IL-4 ($P= 0.81$ and $R= 0.06$) and IL-10 ($P= 0.7$ and $R= 0.10$) in both groups. They experienced no changed during the study period as well ($P<0.05$). Moreover, the study failed to find a significant difference between BMI with plasma concentrations IL-4 ($P= 0.31$ and $R= 0.21$) and IL-10 ($P= 0.15$ and $R= 0.48$), body fat percentage with plasma concentrations of IL-4 ($P= 0.88$ and $R= 0.04$) and IL-10 ($P= 0.74$ and $R= 0.08$) and WHR with plasma concentrations IL-4 ($P= 0.17$ and $R= -0.37$) and IL-10 ($P= 0.78$ and $R= -0.08$) in both groups.

DISCUSSION

The present study demonstrated that, 10 weeks of strength training significantly increased muscular strength, fat-free mass and testosterone and decreased the fat percentage and cortisol. However, the plasma concentration IL-4 and IL-10 and their relationship with testosterone, cortisol and body composition did not change.

In strength group, the increase in the 1RM of the upper and lower body was accompanied by an increase in the fat-free mass of the body which was significantly higher than the control group. This increase in the fat-free body mass indicates that, the strength group exhibited more protein synthesis and as a result experienced greater muscle hypertrophy [26]. The increase in muscle mass the main insulin target site, induced by strength training, not only has positive impact in energy expenditure but also improves insulin sensitivity [12].

The relationship between physical activities and inflammation has been reported in several studies [9, 10, 14]. Lower concentration of inflammatory markers has been observed in individuals who reporting more frequent and more intense physical activities [9, 10]. The reason for this inverse relationship is not fully known, yet it could be concluded that there must be a relationship between physical activity and adiposity. Consequently, it is feasible that body fat has the greatest effect on the concentration of inflammatory markers in circulation [9, 10]. Therefore, it could be concluded that the reason for a lower inflammation in active individuals is primarily because of lower absolute amount of total and visceral body fat [9]. Yet, despite of the 13% decrease in the body fat mass, no significant change was observed in the plasma concentration of the inflammatory cytokines. Moreover, the study failed to find a significant relationship between of the fat mass, or any body composition measures and plasma concentration of cytokines. One group recently demonstrated that, when induced by diet and exercise, weight loss produced cytokine concentration changes that were transient during the period of weight loss, but lost after a subsequent 2-week period of weight maintenance [29]. Thus, since the cytokine changes induced by dieting or medical interventions cause transient weight loss, it could be concluded that cytokine changes are not related to the changes in the fat mass [28]. According to the findings of the present study, alternations in the concentration of cytokine, reported in the past, May be more directly linked to the rate of change in adipose tissue.

Several studies investigating the effects of resistance training on inflammatory markers, have found conflicting results. In one of the first study in 1966, similar to the findings of the present work, a study reported that 12 weeks of incremental resistance training with high intensity does not influence the serum concentration of TNF-α, IL-1β, IL-6 and IL-2 [30]. Similarly, studies by Ferreira et al. (2010) and Levinger et al. (2009) revealed that 10 week circuit resistance training did not induce a change in the serum concentration of pro-inflammatory cytokines and IL-10. Ogawa et al. [31] demonstrated that 12 weeks of resistance training does not change the plasma concentration TNF-α and IL-6 in elderly women. Brochu et al. [19], as well, failed to identify an effect of resistance training without other interventions such as dieting on inflammatory and metabolic parameters.

On the other hand, Balducci et al. [32] showed that a combined resistance and aerobic training for 12 months, reduces the serum concentration IL-4 and IL10 in diabetics and patients with metabolic syndrome. Abd el-Kader et al. [16] reported a decrease in TNF-α and IL-6 in diabetics after a 12-week resistance training program. Reduction in the TNF-α of the elderly women after 10 weeks of training [33] and after 10 months of resistance and flexibility training in male and female patients [1] have also been reported.

The reason for these conflict results from this study and other ones [1, 16, 32, 33] may be the subjects under study, the methodology or the baseline levels of inflammatory markers and study design. Since some studies are conducted on healthy individuals [1, 33] and some others are on patients [16, 30], it could be concluded that studying healthy and young subjects would be easier in order to identify the response inflammatory cytokines have to exercise training. It seems as if the elderly, the obese and the females are more sensitive to the effects of resistance training on inflammatory markers.

Another probable reason for these contradictions may be the intensity and length of the training programs. In the present study, healthy men participated in training programs for only 10 weeks with 70 to 80% maximum repetition but other studies reporting the reduction of inflammatory markers were conducted in a longer periods (more than 10 months) and were more severe [1, 32]. Studies have shown that the length and the intensity of trainings affect the cytokine responses so as the extreme response was reported after a 16 week program with intensities over 80% with one maximum repetition [12]. The intensity of the training and other indices of the protocol, including the number of repetitions produces unique cytokine responses and various adaptations to the exercise training [12, 15].

Furthermore, the intervals in time for samplings may have profound effect on the cytokine responses to resistance training. In the similar studies, sampling was done after 17 [31], 48 [17], and 72 to 96 [1, 8] hours after the last bout. It is reported that, to evaluate the training effects at rest, samples must be taken at least 72 hours after the last bout [12].

The findings of this study demonstrated that despite of the significant 36% reduction in the cortisol (from 146.1 to 109.4 ng/mL) and the 31% increase in testosterone (from 5.1 to 8.8 ng/mL), there was no significant relationship between cytokine changes and plasma concentration of testosterone and cortisol after 10 weeks of strength training.

Since the plasma concentration of the cytokines measured in this study did not have any change, it may be concluded that 10 weeks of training is not sufficient for creating a relationship between changes in the cytokines and cortisol and testosterone and a longer training period would be needed. The present study failed to find a significant relationship between body fat mass, BMI and the waist to hip ratio and the plasma concentration of cytokines. However, there is a probability for further reduction in the percentage of the fat to create more distinct responses by serum concentration of cytokines; however, findings of the present study and some other ones [28, 29] have revealed that the reduction in the fat mass is independent from the systemic changes in inflammatory cytokines.

In conclusion, according to the findings of this study and despite of the significant increase in muscular strength, fat-free mass and testosterone and the decrease in cortisol and body fat mass, 10 weeks of strength training did not induce a significant change in the plasma concentration of anti-inflammatory cytokines in overweight young men. Longer periods of training and combined the strength training with aerobic ones or dieting may have better effects on reducing the concentration of systemic cytokines. These findings also indicated that the changes in the concentrations of systemic cytokines are not directly related to the absolute changes in the fat mass but they are generally related to the fat mass in the body. Testing this hypothesis through controlled interventions may be worthwhile.

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