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ORIGINAL ARTICLE

The effect of eight weeks of aerobic training on the plasma level of adiponectin, leptin, and resistin in healthy middle-aged men

Effets de huit semaines d'entraînement en aérobic sur les concentrations plasmatiques d'adiponectine, leptine et résistine chez des hommes en bonne santé et d'âge mûr

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KEYWORDS Adipokines; Exercise; Untrained men **Summary** Aim of present study was investigating the effects of 8-weeks of aerobic training program on plasma levels of adiponectin and leptin in healthy middle-aged men. *Methods.* – For this purpose, thirty middle-aged healthy men were selected and depended on their body fat percentage were assigned in two equal groups. Experimental group performed 8-weeks aerobic training 4 days a week in alteration days with 60–80 percentage of maximum heart rate and control group remained sedentary. Blood samples were collected prior to and after training program for all subjects and plasma adipokines levels were measured. *Results.* – Results showed a significant increase in adiponectin and resistin concentrations, also a decrease in leptin concentrations following 8-weeks of aerobic training program (P < 0.05). In addition, reduction in body fat percentage (P < 0.05) negatively correlated with adiponectin and resistin levels (P < 0.05). *Conclusion.* – In healthy middle-aged men, 8-weeks of aerobic training reduced body fat percentage and improved plasma adipokines levels, so aerobic training can considered as major strategies for preventing obesity and associated diseases. © 2011 Elsevier Masson SAS. All rights reserved.

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MOTS CLÉS

Adipokines;

Entraînement

Exercice ;

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Résumé

Objectif. — Enquêter sur les effets d'un programme de huit semaines d'entraînement en aérobic sur les niveaux d'adiponectine et de leptine du plasma chez des hommes en bonne santé d'âge mûr.

Patients et méthodes. — Dans ce but, trente hommes en bonne santé d'âge mûr ont été choisis, en fonction de leur pourcentage de masse grasse et divisés en deux groupes égaux. Le groupe expérimental a exécuté huit semaines d'entraînement en aérobic, à raison de quatre jours séparés par semaine, à 60 à 80 % de la fréquence maximale cardiaque et le groupe de contrôle est resté sédentaire. Les échantillons de sang ont été recueillis avant la fin après le programme d'entraînement pour tous les sujets et les niveaux d'adipokine du plasma ont été mesurés.

Résultats. – Ils ont montré une augmentation significative dans les concentrations d'adiponectine et de résistine, et aussi une diminution dans les concentrations de leptine après le programme des huit semaines d'entraînement aérobic (p < 0,05). En plus, la réduction du pourcentage de gras de corps (p < 0,05) a été négativement corrélée avec les niveaux d'adiponectine et les niveaux de résistine (p < 0,05) et de façon positive corrélée avec les niveaux de leptine (p < 0,05). Chez les hommes en bonne santé d'âge mûr, les huit semaines d'entraînement aérobic réduisent le pourcentage de masse grasse corporelle et améliore les niveaux d'adipokines du plasma.

Conclusion. – L'entraînement aérobic peut être considéré comme une stratégie importante pour prévenir la corpulence et la maladie associée.

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1. Introduction

Biologically, fat tissue is more than energy storage, it is rather an active tissue-releasing protein such as adiponectin and leptin [1-4], while other cytokines, such as CRP and resistin, are less released at fat tissues, and are mainly synthesized in immune cells [1,5]. These hormones have autocrine and paracrine effects on the brain, liver, and skeletal muscles. Also they play a crucial role in the diseases caused by obesity through regulating metabolic and inflammatory processes [6].

Adiponectin has anti-inflammation, anti-atherosclerosis, and anti-insulin resistance effects [7]. Adiponectin level has a negative correlation with body fat percentage, central fat distribution and leptin [5,7]. Patients suffering from diabetes, high blood pressure and ischemic heart disease have less adiponectin concentration compared to healthy people [8]. Within four years of study, subjects with hypoadiponectinemia died because of heart diseases more than normal people. Hence, hypoadiponectinemia is introduced as the new cardiac risk factor [9].

Leptin is identified to be of the first adipokines related to the body fat mass and the loss of weight and fat percentage is often accompanied by a decrease in the leptin levels [10,11]. An increase in the level of leptin is observed in cardiovascular, diabetes and even asthmatic patients [12].

Resistin is a hormone belonging to the family of cysteine-rich proteins identified as one of the symptoms of atherosclerosis and is one of the important predicting factors of cardiovascular diseases [13].

The increase in the level of resistin mostly happens in inflammatory conditions and it is shown that resistin stimulates the synthesis and the release of pre-inflammatory cytokines [14].

Reilly et al. have reported that in subjects suffering from metabolic syndrome, the level of resistin in plasma is predictor for coronary atherosclerosis [5].

The high concentration of Adiponectin has negative correlation with insulin resistance. There are findings approving [15] and disapproving [16] of the direct relationship between resistin and insulin resistance and type 2 diabetes.

Changes in plasma adipokine concentrations in response to physical exercise (acute/chronic) has been established in previous studies, but there is lack of researches using healthy subjects.

In the present study, researchers tried to find an answer to this question that, in addition to recognized positive effects on cardiovascular system, whether aerobic training can be effective in changing the amounts of the hormones which play the main role in controlling the metabolic and inflammatory activities of the body and also are the predictors of the diseases such as diabetes and atherosclerosis or not. In order to find out the levels of the hormones of adiponectin, leptin and resistin were measured prior to and after executing exercise program, and their relationship with fat percentage was calculated.

2. Method

2.1. Subjects

The statistical population of this study consisted of 152 middle-aged men, enrolling in a health club. From among them, 30 men were chosen who had never participated in a regular sport activity, no background of disease or taking drugs affecting the concentration of adiponectin, leptin and resistin as to be the subjects. After filling the informed consent form and the form of readiness for participating in sport activities (PAR-Q 2002), subjects were divided into two

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Aerobic Training and Plasma adipokines

Table 1 Baseline des	Baseline descriptive characteristics.					
Variable	Control group (<i>n</i> = 15)	Experimental (n = 15)				
Age	37.06 ± 5.1	40.06 ± 4.04				
BMI	76.22 ± 7.84 24.62 ± 1.99	79.8 ± 8.51 25.67 ± 2.27				
Body fat percentage	$\textbf{18.33} \pm \textbf{2.51}$	$\textbf{18.88} \pm \textbf{2.95}$				
Data are expressed as mean 1 SD						

Data are expressed as mean \pm SD.

equal groups (n = 15) based on their fat percentage. Table 1 shows the individual features of the subjects.

2.2. Training program

The experimental group carried out the aerobic exercises, every other day, four sessions a week, for 8 weeks. These exercises included warm up, main training, and cool down. Subjects warmed up by stretching and jogging for 10 minutes. The main training included basic physical fitness movements (running, jumping and playing with medicine ball), reaching 60 to 80% of the maximum heart rate done for 15 minutes in the first session and increasing gradually to be 35 minutes by the end of the eighth week. Cooling down included the static stretching movements. The performance of the subjects was constantly checked by the polar device (POX 1000 Japan), and control group remained sedentary in this period.

2.3. Controlling nutrition

Since nutrition affects adiponectin, leptin and resistin levels, in order to control the nutrition of the subject, they were asked to write down in a specially designed diary, the quantity and time at which food was eaten in three days before blood sampling. After dietary assessment to determine the total amount of calories for each subject, a dietician prescribed an individualized diet based on 3-days diary form, including the quantity and type of foods to be consumed per day. Three-days before the second blood sampling, subjects follow the designed diet.

2.4. Anthropometric measurement

The subjects were weighted by a digital scale of 0.01 kg accuracy. Fat percentage of the subjects was measured using caliper device by the method of three skinfold thickness (subscapular, abdominal, and triceps) [17].

2.5. Blood sampling and laboratory measurements

Forty-eight hours before starting the exercise program, while all the subjects were fasten; 10 cc of blood was taken from their brachial vein. In order for preventing the data of being interrupted by the circadian rhythm of these three hormones, all blood sampling were done from 8a.m to 9a.m. Also, 48 hours after finishing the 8-weeks exercise program, post-testing blood sampling was done under the same conditions. All the samples were rapidly put in EDTAcontaining tubes and kept in the refrigerator until they were centrifuged. Centrifuge was done at the gravity of 3000, temperature of -4° C, for 15 minutes, and the separated plasma was kept at the temperature of -80° C. Plasma leptin, adiponectin and resistin, were measured using ELISA kits (Mediagnost, Reuttlinger, Germany) for leptin and (Adipogen co., South Korea) for adiponectin. The intra- and interassav coefficients of variation were respectively 3.9, 8.6% for adiponectin, and 3% and 5% for leptin. Plasma resistin was measured by an enzyme-linked immunoassay kit (Ray-Biotech, Inc.). This biotin-labelled antibody sandwich assay measures homodimeric resistin. The intra- and inter-assay CV were 10 and 12%. The probable changes in the volume of plasma after the training program was calculated and the measured amounts of the hormones were corrected based on it [18].

2.6. Statistical analysis

Normality of the data was tested using K-S test and the equality of the variances of the groups in different factors was tested using levene test. After being assured of the normality and equality of the groups, variance analysis test with repeated measuring was used to study the differences between the mean amounts. Also, Pearson test was applied to evaluate the correlation between different variables. Statistic calculations of this study were done by the software SPSS, Ver.16.

3. Results

As expected, after 8 weeks of aerobic exercises, the experiment group showed a significant decrease in weight and fat percentage. The plasma levels of adiponectin and resistin showed an increase and the leptin level showed a decrease in the experimental group, comparing to the control group (Fig. 1 and Table 2).

Also, there was no significant relationship between fat percentage and plasma levels of the three hormones of adiponectin, leptin and resistin (Table 3). However, after studying the percentages of the changes in these variables,



Figure 1 Changes in the fat percentage and the three hormones of adiponectin, leptin and resistin, in experiment and control groups.

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Anthropometric and hormonal indicators of the subjects, before and after the training program.

•		•	••••		
Variable	Control group		Experimental group		
	Pre-test	Post-test	Pre-test	Post-test	
Weight (kg)	79.80 ± 8.51	80.28 ± 8.76	76.29 ± 7.84	75.86 ± 7.54 ^b	
BMI	$\textbf{25.67} \pm \textbf{2.27}$	$\textbf{25.82} \pm \textbf{2.33}$	$\textbf{24.62} \pm \textbf{1.99}$	24.46 ± 1.91^{a}	
Body fat percentage	$\textbf{18.88} \pm \textbf{2.95}$	19.27 ± 3.16	$\textbf{18.33} \pm \textbf{2.51}$	17.86 ± 2.43^{b}	
Adiponectin (μ g/ml)	16.38 ± 1.55	$\textbf{16.29} \pm \textbf{1.47}$	16.75 ± 1.44	18.02 ± 1.84^{b}	
Leptin (ng/ml)	$\textbf{2.28} \pm \textbf{0.21}$	$\textbf{2.33} \pm \textbf{0.19}$	$\textbf{2.25} \pm \textbf{0.24}$	$\rm 2.22\pm0.22^{a}$	
Resistin (ng/ml)	$\textbf{7.36} \pm \textbf{1.28}$	$\textbf{7.31} \pm \textbf{1.18}$	$\textbf{7.14} \pm \textbf{1.32}$	10.21 ± 1.51^{b}	

^a Significance of the changes at the level of 0.05.

 $^{\rm b}$ Significance of the changes at the level of 0.001, as compared to the control group, the results are showed in the form of mean \pm standard deviation.

 Table 3
 The relationship between fat percentage and the three hormones of adiponectin, leptin, and resistin.

Variable	Pre training		Post training	Post training		Change percentage	
	r value	P value	r value	P value	r value	P value	
Adiponectin	0.19	0.29	-0.049	0.79	-0.56	0.001	
Leptin	0.11	0.55	0.19	0.30	0.47	0.008	
Resistin	0.32	0.08	0.00	0.99	-0.63	0.001	

it was showed that the decrease in fat percentage had a negative correlation with the increase of two hormones of adiponectin and resistin, and a positive correlation with leptin.

4. Discussion and conclusion

The present study focused on the adaptation made in the concentration of adiponectin, leptin and resistin, due to training. These are the hormones which appropriate amount of them in the blood, can prevent the diseases related to them, such as diabetes type 2, metabolic syndrome, and cardiovascular diseases.

The results showed that body fat and plasma leptin decreases after 8 weeks of aerobic exercises. This change in the level of leptin has been reported by a great range of previous studies [19-25].

The level of leptin in the blood has reduced after 2 weeks of skiing exercises in a group of men [19]. Also, the similar result was found on serum level of leptin, after a one-year training program in a group of overweight men [20].

The leptin level in a group of old men (65–75 years) divided into three groups of light, moderate and heavy training was decreased after a year of resistance training. Fat percentage and BMI decreased in the three experimental groups and this decrease was more at heavier intensities. The changes in the level of leptin had a direct relationship with the changes in the level of fat, which are in agreement with the results of this study [21].

On the other hand, 6 weeks of strength training [22] and 6 weeks of resistance training [23] did not change the level of leptin of obese subjects. It should be mentioned that the subjects of those study did not show a

significant loss of weight after executing this program. In a temporal-phenomenal historical study on the patients suffering from fatty liver and on healthy people, it was indicated that people, who did aerobic exercises, at least once a week, had lower serum leptin [24].

It seems that in general, long term trainings, which result in a reduction in fat percentage, reduce leptin levels too. The cause of the few contradictions found in the articles may be the lack of a significant weight loss and uncontrolled genetic background of the subjects, where Collins et al. reported that the decrease in the level of leptin because of doing training was higher in healthy people with a family background of diabetes, as compared to the ordinary people [25].

Current study showed that the plasma adiponectin increases following 8 weeks of aerobic training, and it's absolute amounts after increasing are almost two times higher than the patients in the other studies [26] and this high level of adiponectin is probably a preventing factor against diseases related to adiponectin. The changes in the level of adiponectin because of adaptation with the aerobic exercises in most of the previous studies have shown the same results as of this study [27–30] and such agreement is also observed in resistance exercises [31].

In a study on young and middle-aged women, doing ten weeks of aerobic training resulted in an increase in the plasma levels of adiponectin alongside with a decrease in insulin resistance [27].

Also, eight young fat women (BMI \geq 25) showed a significant decrease in the level of fat mass and leptin, also an increase in the level of adiponectin after 7 months of aerobic training program [28].

In another study, a group of fat subjects with insulin resistance were given a program of 19 weeks of aerobic training

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Table 2

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alongside with diet. The results showed that visceral fat was decreased and adiponectin was increased. And also it is reported that adiponectin level is directly related to fat distribution [29], which was not found in present study.

Bruun et al. reported that very obese subjects (mean BMI = 45.8), after executing a program of 15 weeks of aerobic training and a low-calorie diet, showed significant increases in the plasma levels of CRP, IL6 and adiponectin. It can be concluded that diets have a prominent impact on creating increased levels of adiponectin due to exercising [30].

There are also studies, which show exercises to have no effect on the level of adiponectin. That may be because of using a combination of endurance-strength exercises [31-33] or using athlete subjects who have higher adiponectin level in baseline [34] or other unknown factors [24,35].

Kadoglou et al. found that 16 weeks of aerobic training resulted in the reduction in plasma levels of resistin, CRP, and interleukin-6 in the patients suffering from type 2 diabetes [13]. Patients suffering from type 2 diabetes and metabolic syndrome after 12 months of aerobic training showed an increase in their serum level of adiponectin, and also showed decreases in serum levels of resistin and leptin [35].

In another study, nine overweight children performed a progressive aerobic program for 8 weeks. The results showed that none of the adipokines (adiponectin, leptin, resistin, interleukine 6, TNF α and CRP) changed significantly. Researchers announced that the reason for this result is probably the lack of weight loss and that exercising does not directly affect the hormones [36].

As seen, researches which have studied the effect of exercising on people suffering from diseases, mostly has found the resistin to be reduced, while in this study which is done on healthy people who are not overweight, exercising has increased plasma level of resistin. In agreement with the results of this study, Perseghin et al. showed that elite endurance athletes comparing to patients with type 2 diabetes and also healthy subjects with no physical activity have higher levels of resistin and insulin sensitivity [16]. This challenges the direct relationship between resistin level and obesity and associated diseases [15].

Elloumi et al. divided 21 fat teenagers (BMI= 30.8 ± 3.2) into the three groups of low-calorie diet, exercising, and a combination of both. Following 2 months of the protocol of weight loss, the three groups showed an increase in the level of adiponectin and decrease in the level of leptin similar to current study, and that was more significant in the group with the combination of diet and exercise. Also the level of resistin showed a significant increase in the two groups of exercising and combination of exercising and diet [2].

In order to clarify the effect of exercising on the level of resistin in different subjects, a comparative study on ill people, healthy people and athletes seems required. It should be mentioned that in this study, the mean value of increased serum levels of resistin in the experimental group after 8 weeks of practicing was not comparable with the absolute amount of resistin measured in the other patients of the other researches, and it was much lower. It seems that increases in the levels of resistin, which happen in healthy people because of exercising, is not originated by the inflammations resulted from disease, rather they are originated by the inflammation of training. The most important mechanism which can explain the increase in the level of resistin after aerobic exercises in this study, is the role of this hormone in anti-oxidation defences of the body, as Bo et al. have reported in their study that resistin functions as an antioxidant in response to inflammatory stimulant [37].

In conclusion, most important findings were to show that plasma adiponectin, leptin and resistin concentrations will improve followed by 8 weeks of aerobic training that was associated with improvement of body composition. Ideal levels of such adipokines can play an outstanding role in preventing metabolic and cardiovascular disease. Also we reported strong evidence that the resistin concentration increased significantly following 8 weeks of aerobic training in middle-aged healthy subjects that wasn't certainly found in previous studies.

Disclosure of interest

The authors declare that they have no conflicts of interest concerning this article.

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