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Hs-CRP and Adipokin (Lcn2): Response to Exercise Training in Obese Men

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ABSTRACT

Obesity is a condition of excess body fat. It has been known as major public health problem, was reported to be associated with insulin resistance, type 2 diabetes mellitus and cardiovascular disorders. Therefore the purpose of this study was to examine the effects of aerobic training on hs-CRP and adipokin (as Lcn2) in obese young men. Thirty healthy young men (aged 27.83 ± 1.69years, height 1.71± 5.37 cm, BMI 30.1 ± 1.49 kg/m^2, mean ± SD) participated as subjects in this study. The subjects were randomly assigned to aerobic training group (n=15) or control group (n= 15). Aerobic training group underwent an 8-week intervention, with a frequency of 3 d/wk at an intensity corresponding to 65 – 80% maximum heart rate for 35 – 55 min. The results showed that body fat percent, WHR, BMI, were decreased ((P<0.05), in the training group compared with control group. Maximum oxygen consumption, on the other hand, increases significant (P<0.05) in the training group compared with the control group. Adipokin lcn-2, LDL-c, TG, TC decreased (P<0.05) and HDL-c increased (P<0.05). No significant changes in hs-CRP were found after 8 weeks aerobic training. It seems that 8 weeks aerobic training induced change in adipose tissue, decrease Adipokin (Lcn2), but this improvement was not accompanied by decreased hs-CRP in young obese men.

Key words: Aerobic training, Adipokin, hs-CRP, obese men.

INTRODUCTION

Obesity has long been recognized as a major health problem in many countries. It has been attributed to hereditary factors, economic growth, rapid urbanization, and lifestyle changes. It is a serious health problem for increases the risk of developing cardiovascular diseases, type2 diabetes, hyperlipidemia, hypertension, and increased mortality (Yang, 2005). It's also an increase in fasting glucose, blood pressure, triglycerides and a decrease in high density lipoprotein (HDL) So it's known as metabolic syndrome (Wang, 2007). Studies have demonstrated close associations between obesity and increased circulating concentrations of proinflammatory molecules, including acute-phase proteins, cytokines, adipokines, and chemokines (Tataranni. 2005; Weisberg, 2003). In obese states, these proinflammatory factors are produced predominantly from enlarged adipocytes and activated macrophages in adipose tissue and liver. Many of these inflammatory factors, such as interleukin-6 (IL-6), tumor necrosis factor-α (TNF-α) and hs-CRP, can directly induce glucose intolerance and insulin resistance by antagonizing insulin's metabolic actions at peripheral tissues, especially in liver and skeletal muscle (Fantuzzi, G. 2005). C-reactive protein (CRP) is produced by the liver and its role in the development of inflammation has been well established. However, the strong association between CRP and risk for heart disease and obesity is a more recent discovery, in other hand, there is a moderate to strong positive correlation between CRP and body mass index (BMI) reported by numerous population-based studies (Patrica HU, 2008). Adipokin Lipocalin2 (Lcn2) also known as

neutrophil gelatinase associated lipocalin, sidrocalin and 24p3, is another member of the lipocalin family recently reported to have possible metabolic roles. (Fantuzzi, G. 2005). Lipocalin2 is expressed in many tissue, including neutrophils, macrophage, kidney, liver, lung, thymus, small intestine mammary tissue as well as adipocytes and is known to play a role in inflammation. Lcn2 has been recognized as an adipocyte drive acute phase protein that is positively correlated with potential effect in obesity inflammation and insulin resistance in mice and humans (Choi. 2009; Wang, 2007). It also has been showing that circulating levels of this adipokin has a strong direct correlation with hs-CRP as an acute phase protein (Van Dam. 2007). The lack of physical activity in daily life induces obesity and increases the risk of hypokinetic diseases; diabetes mellitus, hypertension, heart diseases etc. Exercise is well known as the cornerstone treatment for obesity-related metabolic complications, including insulin resistance, hypertension, impaired glucose tolerance or diabetes, hyperinsulinemia, and dyslipidemia, that are characterized by elevated adipose accumulation (Hu, 2001; Tuomilehto. 2001).Therefore one of the best strategies for preventing obesity and its associated inflammation is participation in regular physical activity (Petersen & Pedersen, 2005). On the other hand, exercise has been shown to have beneficial effects on obesity, type2 diabetes and the metabolic syndrome. Although the changes in adipokine levels might be an important clue for understanding the beneficial effects of exercise, data on exercise-induced changes of inflammation factors such as adipokin and hs-CRP…is still unclear. Recently Dabidi Roshan, (2011) reported that aerobic exercise training has positive influence to concentrations of hs-CRP and blood lipids in Rats. Patricia CH et al. (2008) isn't reported that any changes in CRP level in obese male adolescents after 12 weeks exercise training. Recently, Damirchi (2011) reported that Lcn2 increased after single bout graded exercise in obese and normal weight men. Choi et al (2009) in an only available study, isn't reported that any change in Lcn2 level in obese women after 12 weeks moderate exercise training. The physiological and biochemical responses to resistance exercise are different from those exhibited in response to aerobic exercise (Kraemer, 1994). No previous study has investigated the effects of aerobic exercise on hs-CRP and Lcn2 concentration in obese young men. Therefore the present study was designed to determine the effects of aerobic training on hs-CRP, Adipokin Lcn2 and other inflammation factors.

MATERIAL AND METHODS

Subjects

Thirty healthy and university students aged (aged 27.83 ± 1.69years, mean±SD) enrolled in this study .The inclusion criteria were men who had body mass index (BMI) ≥29.9 kg/m^2 did not engage in regular exercise training at the time of their enrolment. Student who were afflicted with heart diseases, hypertension, pulmonary diseases and diabetes, who needed orthopedic treatment, and who had neurological limitations to physical exercise were excluded. All the subjects were asked to complete a personal health and medical history questionnaire, which served as a screening tool. The subjects were given both verbal and written instruction outlining the experimental procedure, and written informed consent was obtained. All the subjects completed the 3-day diet recall forms and were instructed to maintain their normal physical activity and dietary habits throughout the study. The subjects were randomly assigned to one of the experience group (n=15) and control group (n=15).

Exercise training

The participant's of experience group (aerobic training) underwent three exercise training sessions per week for 8weeks. The training exercise consisted of a 10-minute warm-up period, as well as muscle stretches. It's also consisted of walking and running at 65-80% of maximal heart rate (HR_{max}) for 35-55 min per day, 3 days per week, for 8 weeks. The programme started with 30 min running for the first few sessions, and this was then changed to 45 min per session until the end of training. Each training session finished with a cool down. The exercise intensity was controlled by the authors, using a hear rate monitor, who ensured that it was between 65 and 80% of HR_{max} throughout the trial.

Measurements: (Anthropometric and body composition measurements)

Height and body weight were measured, and body mass index (BMI; kg/m2) was calculated from height and weight of each subject. Waist

circumference was determined by obtaining the minimum circumference (narrowest part of the torso, above the umbilicus) and the maximum hip circumference while standing with their heels together. The waist to hip ratio (WHR) was calculated by dividing waist by hip circumference (cm) (ACSM , 2005). Subcutaneous body fat was measured at 3 sites (chest, abdominal, and thigh) with a Lafayette caliper. Body fat percent was calculated from the formula developed by Jackson and Pollock (1985) .VO_{2max} was determined by Rockport One-Mile fitness walking test. In this test, an individual walked 1 mile (1.6 km) as fast as possible on a track surface. Total time was recorded and HR was obtained in the final minute (ACSM) [1]. VO_{2max} was calculated by following formula:

$VO_{2max =}$ [139.68-(0.388×age (year))] - [0.077× body mass (pb)] - [3.265×time (min)] - [0.156× HR].

Biochemical analyses

Approximately 10 milliliters of blood was collected into plain and EDTA filled vacutainer tubes after an overnight fast of at least 12 hours at the same time before and after 8 weeks intervention. The tubes were then centrifuged and serum and plasma were drawn off and stored at -80°C until analysis. hs-CRP levels were determined in duplicate via an ELISA kits (Diagnostics Biochem Inc, Canada). The intra and inter-assay coefficients of variation for hs-CRP were <5.7% and a sensitivity of 10 ng/ml. The adipokin Lcn2 level was measured in duplicate using an enzyme-linked immunosorbent assay (ELISA) kits (Uscn Life Science Inc, Wuhan, China). The sensitivity of kit was 0.12 ng/ml. Serum cholesterol, triglycerides, HDL-c and LDL-c were assayed with automated techniques.

Statistical analysis

Statistical analyses were performed with SPSS program (version 16, SPSS, Inc., Chicago, IL). Values were expressed as mean ± standard deviation (SD). Independent t-test and paired t-test were used to evaluate changes in variables. General linear regression analysis and Pearson's correlation were performed to calculate a correlation between variables in response to training. P-values less than 0.05 were considered statistically significant.

RESULTS

Anthropometric, physiological and metabolic characteristics of subjects are shown in Table *1 and 2*. The results showed that body weight, body mass index (BMI), body fat percent and WHR were decreased (P<0.05) after aerobic training. Maximum oxygen consumption, on the other hand, increases significant (P<0.05) in the training group compared with the control group. Plasma lipocalin-2, LDL-c,TG, TC decreased (P<0.05) and HDL-c increased (P<0.05) after 8 weeks aerobic training (Table 1).For hs-CRP, there was no significant different between aerobic training group and control group after 8 weeks exercise. Pearson's correlation demonstrated a positive relationship between, adipokin (Lcn-2) levels at baseline (P<0.05) with body fat percent, WHR and BMI. No significant relationship between hs-CRP with biochemical variables were found in the endurance group after 8 weeks intervention.

DISCUSSION

CRP is a key inflammatory factor produced by the liver in response to an acute infection or inflammation and its concentration in plasma can increase as much as 1000-fold during injury and infection (Schultz .1990). Adipokin (Lcn2) has been identified as a novel adipokine associated with obesity, type2 diabetes and the metabolic syndrome. The effects of aerobic training on hs-CRP and plasma Lcn2 are still unclear, thus this study aimed to investigate the effects of aerobic training on this factors in obese young men. In this study, no significant differences were observed in hs-CRP after 8 weeks aerobic training, although no previous study has investigated the effects of aerobic exercise on hs-CRP concentration, Mattusch F *et al*., (2000) indicated th**e r**eduction of the plasma concentration of C-reactive protein following nine months of endurance training. These findings demonstrated the possibility of improving the status of CRP with long-term exercise intervention. However in this study, we did not detect positive changes in CRP concentrations following 8 weeks of exercise training programme, which could be considered as a short-term programme (Summerbell.2005). On the other hand, adipokin Lcn2 can be recognized as an inflammatory marker that increases after a progressive physiological stress in sedentary individual. Furthermore, increasing of Lcn2 secretion from fat cells may be stimulated by lip polysaccharides that suggesting

Lcn2 as an acute phase protein (Wang.2007). Results showed that Plasma Lipocalin-2 decreased (P<0.05, 11.2%) in response to 8 weeks aerobic training compared to the control group. Choi (2009) indicated that there was no significant change in the Lcn2 in obese women after 12 weeks moderate exercise training. This discrepant result may be attributed to variation in the exercise protocols and differences in subject. On the other hand, there was the positive relationship between plasma lipocalin2 and body fat percent at baseline populations. The results showed that body weight; body mass index (BMI), body fat percent and WHR were decreased after aerobic training, thus exercise-induced changes in body fat, especially visceral adipose tissue, may attribute to plasma Lipocalin2 decrease and after the training. The results are in agreement with previous reports showing that there was a significant positive relationship between plasma lcn2 levels with body mass, body fat percentage and WHR, suggesting that the increased fat mass might account for the elevated blood levels of this adipokine in obese individuals. Wang (2007), showed a higher concentration of Lcn2 in obesity and this adipokine is positively related to the BMI, Waist circumference and body fat percentage. Choi et al

Table 1: Physical and physiological characteristics of the subjects before and after training (mean ± SD)

Variables	Control		Aerobic Training	
	Pre-test	Post-test	Pre-test	Post-test
Body weight (kg)	81.30 ± 6.76	81.29 ± 6.40	83.65 ± 7.04	80.90 ± 6.77 *
BMI (kg/m^2)	28.49 ± 10.45	28.50 ±10.43	28.67 ± 1.50	27.67 ± 1.41 *
%Body fat	23.56 ± 1.53	23.36 ± 1.65	23.66 ± 2.22	22.48 ± 2.0 *
WHR	.90 ± .03	.90 ± .03	.93 ± .02	.89 ± .03 *
VO_{2max} ($ml.kg^{-1}.min^{-1}$)	35.76 ± 3.37	35.96 ± 3.23	35.9 ± 2.77	43.68 ± 2.39*†

* P<0.05 for between-group differences.

† P<0.05, pretraining *vs.* posttraining values.

Table 2: Metabolic characteristics of the subjects before and after training

Variables	Control group		Training(Endurance group)	
	Pre-test	Post-test	Pre-test	Post-test
Triglyceride (mg/dl)	1.61± 2.03	1.58 ± 2.03	1.71± 4.56	1.48 ± 4.41
Cholesterol (mg/dl)	1.89 ± 2.7	1.88 ± 2.6	1.96 ± 4.22	1.85 ± 3.33
LDL-c	1.27 ± 2.57	1.27 ± 2.39	1.32 ± 3.1	1.24 ± 2.74 *
HDL-c	38.01 ± 4.98	38.23 ± 5.27	35.4 ± 6.81	42.1 ± 6.51 *
Adipokin(Lcn2(ng/ml)	23.56 ± 2.26	23.02± 2.8	23.79 ± 2.82	19.33 ± 2.45 *†
Hs-CRP	.83 ± .88	.86 ± .88	.78 ± .73	.72 ± .55

* P<0.05 for between-group differences.

† P<0.05, pretraining *vs.* posttraining values.

(2008) demonstrated that a positive relationship between Lcn2 and body mass and Damirchi et al. (2011) showed a positive relationship between Lcn2 level with waist circumference, fat mass and BMI. Body fat percent decreased 8.8% after 8 weeks endurance training, thus it seems that the aerobic training could offer a sufficient stimulus for plasma Lcn2 decreases .Results showing no significant relationship between Lcn2 and hs-CRP after 8 weeks aerobic training. Suggesting that decrease of the other inflammatory markers might decrease Lcn2 and CRP concentration. Serum CRP levels

correlated with serum IL-6 and TNF-α concentration in this study, which then affects the production of CRP by the liver. Additional research is needed to examine whether exercise induced change in IL-6 and TNF-α concentrations, decreases CRP. We did not measure IL-6 and TNF-α in the present study. If we could measure these inflammatory markers, we could carefully explain the decrease of plasma Lcn2 in response to 8 weeks exercise training in obese men.

CONCLUSIONS

In conclusion, aerobic training induced change in adipose tissue, decrease plasma lipocalin2 in obese men. These findings suggested that changes in adipokin lcn2 levels may be associated with the beneficial effect of exercise. Further studies are needed to elucidate the mechanisms responsible for the effects of exercise on CRP and adipokines.

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REFERENCES

1. ACSM., Guidelines for exercise testing and prescription. Philadelphia: Lippincott Williams & Wilkins, pp 57-90 (2005).
2. Choi, K. M., Kim T. N., Yoo, H. J., Lee, K. W., Cho, G. J., Hwang, T. G., Baik, S. H., Choi, D. S., Kim, S. M. Effect of exercise training on A-FABP, Lipocalin-2 and RBP4 levels in obese women. *Clinical Endocrinology*, **70**: 569-574 (2009).
3. Choi, K.M., Lee, J.S., Kim, E.J., Baik, S.H., Seo, H.S., Choi, D.S., Oh, D.J. & Park, C.G. Implication of lipocalin-2 and visfatin levels in patients with coronary heart disease. *Eur J Endocrinol* **158**: 203-207 (2008).
4. Damirchi A, Rahmaninia F,Mehrabani J. Lipocalin2: araesponse to a short tredmill protocol in obese and normal weight men. *Journal of human sport & Exercise.* **6**(1): 59-66 (2011).
5. Fantuzzi, G., Adipose tissue, adipokines and inflammation. *Journal of Allergy and Clinical Immunology*, **115**: 911-919 (2005).
6. Jackson, A.S., Pollock, M.L. Practical assessment of body composition. *Physician Sportsmed*. 76-90 (1985).
7. Kraemer, W. J., General adaptations to resistance and endurance training programs. In Essentials of Sreength Training and Conditioning. TR Baechle, Champaign, IL: Human Kinetics, 127-150 (1994).
8. Peterson, A. M. & Pederson, B. K., The anti-inflammmatory effect of exercise. *Journal of Applied Physiology*, **98**: 1154-1162 (2005).
9. Tataranni,P. A. & Ortega, E.A., Burning question: Does an adipokine induced activation of the immune system mediate the effect of overnutration on type 2 diabetes*? Diabetes*, **54**: 917-927 (2005).
10. Van Dam, R. M. & Hu, F. B., Lipocalin and Insulin resistance: etiological role of retinol-binding protein4 and lipocalin2? *Clinical Chemistry*, 53- 57 (2007).
11. Wang y, Lam KS, Kraegen EW, Sweeney G, Zhang J, Tso AW, *et al*. Lipocalin-2 is an inflammatory marker closely associated with obesity, insulin resistance and hyperglycemia in humans. *Clinical chemistry.* **53**: 34-41 (2007).
12. Weisberg, S. P., McCann, D., Desai. M., Rosenbaum, M., Leibel, R.L. & Ferrante, A.W. Jr., Obesity is associated with macrophage accumulation in adipose tissue. *Journal of Clinical Investigation*, **112**: 1796-1808 (2003).
13. Yan QW, Yang Q, Mody N, Graham TE, Hsu CH, Xu Z, et al.The adipokine lipocalin 2 is regulated by obesity and promotes insulin resistance. *Diabetes.* **56**: 2533–40 (2007).
14. Schultz DR, Arnold PI. Properties of four acute phase proteins: C-reactive protein, serum amyloid A protein, a1-acid glycoprotein

and fibrinogen. *Semin Arthritis Rheum.* **20**: 129-47 (1990).

15. Mattusch F, Dufaux B, Heine O, Mertens I, Rost R. Reduction of the plasma concentration of C-reactive protein following nine months of endurance training. *Int J Sports Med* **21**: 21-4 (2000).
16. Summerbell CD, Waters E, Edmunds LD, Kelly S, Brown T, Campbell KJ. Interventions for preventing obesity in children. *Cochrane Database Syst Rev* **2**: CD001871 (2005).
17. Hu FB, Manson JE, Stampfer MJ, Colditz G, Liu S, Solomon CG, Willett WC**.** Diet, lifestyle, and the risk of type 2 diabetes mellitus in women. *N Engl J Med* **345**: 790-797 (2001).
18. Tuomilehto J, Lindstro¨m J, Eriksson JG, Valle TT, Ha¨ma¨la¨inen H, Ilanne-Parikka P, Keina¨nen-Kiukaanniemi S, Laakso M, Louheranta A, Rastas M, Salminen V, Uusitupa M; Finnish Diabetes Prevention Study Group**.** Prevention of type 2 diabetes mellitus by hanges in lifestyle among subjects with impaired glucose tolerance. *N Engl J Med* **344**: 1343-1350 (2001).
19. Yang, S.Q., "Cytokines and the pathogenesis of non-alcoholic steatohepatitis", *Gut*, **54**: 303-306 (2005).
20. Wang, B., Wood, I. S., & Trayhurn, P., Dysregulation of the expression and secretion of inflammation related adipokines by hypoxia in human adipocytes. *Pflugers Archives,* **455**: 479-492 (2007).
21. Dabidi Roshan V*, "*Effect of Training on High Sensitive C - reactive protein and Blood Lipids Responses in Rats" *Middle-East Journal of Scientific Research* **9**(1): 115-122 (2011).
22. Patricia CH Wong, *et al.*, *Journal of Annals Academy of Medicine*."Effects of a 12-week Exercise Training Programme on Aerobic Fitness, Body Composition, Blood Lipids and C - reactive protein in Adolescents with Obesity" **37**(4): (2008).