Available online at www.sciencedirect.com
ScienceDirect
Metabolism Clinical and Experimental 60 (2011) 186 – 194
Metabolism
Clinical and Experimental
www.metabolismjournal.com

Influence of acute aerobic exercise on adiponectin oligomer concentrations in middle-aged abdominally obese men

Shigeharu Numao[a,*], Yasutomi Katayama[b], Yoichi Hayashi[c], Tomoaki Matsuo[d], Kiyoji Tanaka[d]

[a]*Faculty of Sport Sciences, Waseda University, Tokorozawa, Saitama 359-1192, Japan*
[b]*Faculty of Education, Kogakkan University, Ise, Mie 516-8555, Japan*
[c]*Faculty of Engineering Education Center, Chiba Institute of Technology, Narashino, Chiba 275-0023, Japan*
[d]*Graduate School of Comprehensive Human Sciences, University of Tsukuba, Tsukuba, Ibaraki 305-8577, Japan*

Received 3 June 2009; accepted 9 December 2009

Abstract

Exercise intensity may induce changes in total adiponectin and adiponectin oligomer levels. However, the effects of acute aerobic exercise on total adiponectin and adiponectin oligomers in middle-aged abdominally obese men remain unknown. The purpose of this study was to investigate the influence of aerobic exercise intensity on changes in the concentrations of total adiponectin and adiponectin oligomers (high–molecular weight [HMW] and middle– plus low–molecular weight [MLMW] adiponectin), and the endocrine mechanisms involved in exercise-induced changes in adiponectin oligomer profiles in middle-aged abdominally obese men. Using a crossover design, 9 middle-aged abdominally obese men (age, 54.1 ± 2.4 years; body mass index, 27.9 ± 0.6 kg/m^2) underwent 2 trials that consisted of 60 minutes of stationary cycle exercise at either moderate-intensity (ME) or high-intensity (HE) aerobic exercise (50% or 70% of peak oxygen uptake, respectively). Blood samples were collected to measure the concentrations of adiponectin oligomers, hormones (catecholamines, insulin, and growth hormone), metabolites (free fatty acid, glycerol, triglyceride, and glucose), and cytokines (interleukin-6 and tumor necrosis factor–α). After exercise, plasma catecholamine concentrations were higher during HE than during ME ($P < .05$). Total adiponectin concentration decreased at the end of HE ($P < .05$), but remained unchanged after ME. The HMW adiponectin concentration did not change at either intensity, whereas the MLMW concentration decreased at the end of HE ($P < .05$). The ratio of HMW to total adiponectin concentration increased significantly ($P < .05$), whereas the ratio of MLMW to total adiponectin concentration decreased significantly ($P < .05$), at the end of HE. The percentage changes in epinephrine concentration from baseline to the end of exercise were correlated with the percentage changes in total adiponectin concentration ($r = -0.67$, $P < .05$) and MLMW adiponectin concentration ($r = -0.82$, $P < .05$) from baseline to the end of HE. Our results indicate that the change in total adiponectin was mainly due to a change in MLMW adiponectin concentration during high-intensity exercise in middle-aged abdominally obese men. Epinephrine may partially regulate the decrease in total and MLMW adiponectin concentrations during high-intensity exercise.

1. Introduction

Adiponectin is an adipokine consisting of 244 amino acids that is produced and secreted by adipose tissue. It is highly concentrated (3-30 μg/mL) in circulating blood in humans [1]. The adiponectin concentration is inversely related to metabolic disorders such as metabolic syndrome [2], myocardial infarction [3], dyslipidemia [2], and insulin resistance [4,5]. Adiponectin in blood combines through its collagen domain to form 3 major oligomers: trimer (low–molecular weight [LMW]), hexamer (middle–molecular weight [MMW]), and 12-18-mer (high–molecular weight [HMW]) adiponectin [6,7]. High–molecular weight adiponectin is believed to be the most biologically active form of the 3 oligomers [6-8]. High–molecular weight adiponectin exhibits the greatest binding activity to membrane fractions and activates adenosine monophosphate (AMP)–activated protein kinase most potently of the 3 oligomers in C2C12 myocytes [8]. In humans, several studies reported that the

Institutional approval: This study was approved by the Comprehensive Human Sciences Review Board at the University of Tsukuba.

* Corresponding author.
E-mail address: numashige@aoni.waseda.jp (S. Numao).

0026-0495/$ – see front matter © 2011 Elsevier Inc. All rights reserved.
doi:10.1016/j.metabol.2009.12.011

blood HMW adiponectin concentration is a better predictor for metabolic syndrome [9-11], insulin resistance [9,11], and coronary artery disease [12] than the total adiponectin concentration or LMW adiponectin. These findings suggest that HMW adiponectin, rather than MMW and LMW adiponectin, plays important roles in glucose and lipid metabolism in skeletal muscle [8] and liver [6].

Aerobic exercise is recommended to prevent and improve metabolic abnormalities [13]. Acute aerobic exercise and chronic aerobic exercise improve glucose and lipid metabolism [13]. Because adiponectin influences glucose and lipid metabolism, acute aerobic exercise may alter blood adiponectin concentration. Several studies have investigated changes in blood adiponectin concentrations during acute aerobic exercise. Jürimae et al [14] reported that the plasma total adiponectin concentration decreased significantly below the resting values after high-intensity exercise (6000-m time trial using a rowing ergometer, ~20 minutes' duration) in trained men. They also found that the plasma total adiponectin concentration decreased after acute high-intensity exercise (2000-m single sculling, ~7 minutes) after 24-week training [15]. Nevertheless, most studies have demonstrated that the plasma or serum total adiponectin concentrations are unchanged during acute aerobic exercise at various intensities (50% maximum oxygen consumption [$V_{O_{2max}}$] to exhaustion) in healthy individuals [16-21]. Adiponectin oligomer concentrations, such as LMW, MMW, and HMW adiponectin concentration, do not change during acute aerobic exercise in healthy individuals [19,21]. These studies suggest that acute aerobic exercise, at various intensities, induces little change in the concentration of total adiponectin and adiponectin oligomers in healthy individuals.

To the best of our knowledge, little is known about the effect of acute aerobic exercise on adiponectin concentrations in overweight or obese individuals. Recently, Højbjerre et al [20] found that the plasma adiponectin concentration does not change in young overweight men during moderate-intensity (55% $V_{O_{2max}}$) acute aerobic exercise; however, they did not observe changes in the concentrations of adiponectin oligomers. The effect of exercise intensity on the concentration of adiponectin and its oligomers in overweight or obese individuals is also unknown. Exercise intensity affects the kinetics of hormones and metabolites [22,23]. In particular, catecholamines, which increase with exercise intensity, may inhibit adiponectin gene expression [24-28] and secretion in adipose tissue [25-28]. Interleukin-6 (IL-6) and tumor necrosis factor–α (TNFα) are also potent inhibitors of adiponectin gene expression and secretion [29,30] in adipose tissue. The concentration of IL-6 increases during exercise; and the magnitude of the increase is related to exercise intensity [31], whereas the changes in TNFα concentration are relatively small [20,32]. For the reasons outlined above, increased exercise intensity may decrease the concentration of adiponectin and its oligomers during aerobic exercise.

The purpose of this study was to investigate (*a*) the effect of acute aerobic exercise on the concentrations of adiponectin and its oligomer and (*b*) the association between adiponectin concentrations, hormones, and metabolites during moderate- and high-intensity aerobic exercise in middle-aged abdominally obese men. We hypothesized that changes in hormone and cytokine levels induced by increasing exercise intensity would decrease the concentration of adiponectin and its oligomers during aerobic exercise.

2. Methods

2.1. Participants

Nine untrained (no regular exercise training), middle-aged, abdominally obese men (age, 54.1 ± 2.4 years) participated in this study (Table 1). Abdominal obesity was defined as body mass index (BMI) greater than 25 kg/m^2 and waist circumference greater than 85 cm according to the criteria used in Japan [33]. The participants had no history of any metabolic and cardiovascular diseases, but 3 participants were suspected of showing insulin resistance (homeostasis model of assessment for insulin resistance was >2.5). None of the participants were smokers, and all refrained from taking any medications or supplements known to affect metabolism. Their body mass was stable (<3-kg change in body mass) for at least 2 months. The purpose, design, and risks of the study were explained to each participant; and each participant provided written informed consent. The study conformed to the principles outlined in the Helsinki Declaration and was approved by the Graduate School of Comprehensive Human Sciences Review Board at the University of Tsukuba.

2.2. Preliminary measurements

Participants initially performed an incremental exercise test to exhaustion on a cycle ergometer (818E; Monark, Stockholm, Sweden) to determine peak oxygen uptake (V_{O2}peak). During the test, gas exchange (oxygen uptake and carbon dioxide production) was measured by indirect calorimetry (Oxycon α; Mijnhardt, Bunnik, Netherlands); and heart rate (HR) was monitored by electrocardiography

Table 1
Characteristics of participants (n = 9)

Age (y)	54.1 ± 2.4
Height (cm)	167.6 ± 1.7
Weight (kg)	78.7 ± 2.8
BMI (kg/m^2)	27.9 ± 0.6
Waist circumference (cm)	97.3 ± 5.5
%Fat (%)	32.3 ± 0.9
Fat mass (kg)	25.3 ± 0.9
Lean tissue mass (kg)	51.2 ± 2.2
$V_{O_{2peak}}$ (mL/kg/min)	30.6 ± 1.7
HOMA-IR	2.4 ± 0.3

Values arc means ± SE. HOMA-IR indicates homeostasis model assessment of insulin resistance.

(Dyna Scope; Fukuda Denshi, Tokyo, Japan). Peak oxygen uptake was determined according to methods described by Tanaka et al [34].

2.3. Body composition

Height and body mass were measured using a wall-mounted stadiometer and a digital scale, respectively. Body mass index was calculated as body mass (in kilograms) divided by height squared (in square meters). Waist circumference was measured at the level of the umbilicus using a nonelastic tape to the nearest 0.1 cm.

Whole-body fat mass and lean tissue mass were measured by dual-energy x-ray absorptiometry (Lunar DPX-L densitometer; Lunar, Madison, WI) [35].

2.4. Activity and diet before exercise

All participants were instructed to maintain their usual daily dietary and physical activity patterns during the entire study. Participants were instructed to refrain from transient strenuous physical activity for at least 3 days before each trial. On the day before each trial, they kept a food diary and consumed the same meal before each exercise trial.

2.5. Experimental design

Each participant performed 2 trials: a moderate-intensity aerobic exercise trial (ME) and a high-intensity aerobic exercise trial (HE), separated by at least 1 week. The ME and HE consisted of 60 minutes of aerobic exercise at an intensity of 50% or 70% $V_{O_{2peak}}$, respectively. During each trial, gas exchange was measured continuously to estimate total energy expenditure and fat oxidation based on oxygen uptake, carbon dioxide production, and respiratory exchange ratio (RER). Blood samples were collected at rest, 30 minutes, and 60 minutes of exercise to evaluate concentrations of plasma epinephrine, norepinephrine, glucose, serum total and HMW adiponectin, insulin, growth hormone, free fatty acids (FFA), glycerol, triglycerides, IL-6, and TNFα.

2.6. Experimental protocol

The ME and HE were performed in a random order. After an overnight fast for at least 12 hours, participants arrived at the laboratory at the University of Tsukuba at 8:00 AM. After resting for 30 minutes, a resting blood sample (14 mL per sample) was taken. Each trial started at 8:30 AM for all participants. For ME and HE, participants performed cycling exercise for 60 minutes with a workload corresponding to 50% and 70% $V_{O_{2peak}}$, respectively. Gas exchange was measured during both trials using indirect calorimetry. Fat oxidation rates were calculated from RER [36]. Energy expenditure and substrate oxidation were calculated every 10 minutes. Total energy expenditure and substrate oxidation were defined as the sum of energy expenditure and substrate utilization, respectively. Gas analyzers were calibrated before each trial. Heart rate was monitored continuously during the trials and recorded every 5 minutes. Blood samples were collected from each participant before exercise, 30 minutes after starting exercise, and immediately after completing exercise.

2.7. Blood sampling

Blood samples were collected in an 8-mL tube containing thrombin and a heparin-neutralizing agent, a 7-mL tube containing EDTA-Na_2, and a 2-mL tube containing EDTA-Na_2, heparin-Na, and sodium fluoride. All tubes were centrifuged, and plasma and serum were stored at −80°C until analysis. These samples were analyzed to determine concentrations of serum total adiponectin, HMW adiponectin, FFA, glycerol, insulin, growth hormone, triglycerides, IL-6, and TNFα concentrations, and plasma epinephrine, norepinephrine, and glucose concentration. Hemoglobin and hematocrit values were measured to assess plasma volume changes [37].

2.8. Blood analysis

Plasma epinephrine and norepinephrine were quantified by high-performance liquid chromatography (Tosoh, Tokyo, Japan) (epinephrine—assay sensitivity, 54.6 mol/L; interassay coefficient of variation [CV], <5.3%; intraassay CV, <1.0%; norepinephrine—assay sensitivity, 0.06 nmol/L; interassay CV, <5.3%; intraassay CV, <0.3%). The serum insulin concentration was measured by an electrochemiluminescence immunoassay (Roche Diagnostics, Tokyo, Japan) (assay sensitivity, 1.2 pmol/L; interassay CV, <2.8%; intraassay CV, <1.3%). The serum growth hormone concentration was measured with an immunoradiometric assay (SRL, Tokyo, Japan) (assay sensitivity, 1.2 μg/mL; interassay CV, <5.9%; intraassay CV, <5.6%). The serum FFA concentration was analyzed by an enzymatic colorimetric method (Wako Pure Chemical Industries, Osaka, Japan) (assay sensitivity, 0.01 mmol/L; interassay CV, <1.3%; intraassay CV, <0.4%). The serum glycerol concentration was analyzed by an enzymatic colorimetric method (Wako Pure Chemical Industries) (interassay CV, <5.0%; intraassay CV, <5.0%). The serum triglyceride concentration was analyzed by an enzymatic colorimetric method with a glycerol blank sample (Kyowa Medex, Tokyo, Japan) (assay sensitivity, 0.01 mmol/L; interassay CV, <2.3%; intraassay CV, <0.8%). The plasma glucose concentration was analyzed using a glucose oxidase–immobilized membrane–oxygen electrode (Wako Pure Chemical Industries) (assay sensitivity, 0.1 mmol/L; interassay CV, <0.7%; intraassay CV, <0.3%). The serum IL-6 and TNFα concentrations were measured by enzyme-linked immunosorbent assays (R&D systems, Minneapolis, MN) (IL-6—assay sensitivity, 0.7 pg/mL; interassay CV, <6.4%; intraassay CV, <4.2%; TNFα—assay sensitivity, 0.5 pg/mL; interassay CV, <7.4%; intraassay CV, <5.2%). The total and HMW adiponectin concentrations were measured by enzyme-linked immunosorbent assay (Sekisui Medical, Tokyo, Japan) (assay sensitivity, 0.038 ng/mL; interassay CV,

<6.0%; intraassay CV, <6.0%) [38]. The middle– plus low–molecular weight (MLMW) adiponectin concentration was calculated as the difference between concentrations of total adiponectin and HWM adiponectin.

2.9. Statistical analysis

All data are expressed as mean ± SE. To compare changes in hormones, metabolites, IL-6, TNFα, and total, HMW, and MLMW adiponectin concentrations between trials over time, 2-way repeated-measures analysis of variance was used with trial and time as factors. Where appropriate, Tukey honestly significant difference post hoc test was performed to determine specific significant differences over time within trials. To compare the differences in exercise intensity, RER and energy expenditure during aerobic exercise were compared between ME and HE using paired *t* tests. Spearman correlation coefficients were used to determine the associations between change in concentrations of adiponectin and its oligomers, and change in concentrations of hormones, metabolites, and cytokines. There were no significant differences in plasma volume changes between ME and HE, and the measurement values are not adjusted. Statistical significance was set at a confidence level of .05.

3. Results

3.1. Exercise intensity and energy expenditure

Mean HR during ME and HE was 107 ± 5 and 128 ± 3 beats per minute, respectively (Table 2), representing 63% ± 3% and 76% ± 1% of the maximum HR. Mean relative exercise intensity (percentage $V_{O_{2peak}}$) during ME and HE was 50% ± 1% and 69% ± 2%, respectively. Total energy expenditure and fat oxidation estimated by gas exchange were 1529 ± 139 and 761 ± 77 kJ during ME and 2117 ± 136 and 633 ± 108 kJ during HE, respectively.

3.2. Plasma hormone responses

Significant trial × time interactions were found for changes in plasma epinephrine and norepinephrine ($P < .05$) (Table 3). The plasma epinephrine concentration increased significantly from rest to 30 and 60 minutes during HE ($P < .05$). The plasma epinephrine concentration at 30 and 60 minutes was higher during HE than during ME ($P < .05$). The plasma norepinephrine concentration increased during ME and HE ($P < .05$) and was significantly higher at 30 and 60 minutes during HE than during ME ($P < .05$). The serum insulin concentration decreased significantly during ME and HE ($P < .05$). No significant trial × time interaction was found for changes in serum insulin concentration. The serum growth hormone increased significantly during HE ($P < .05$).

Table 2
The relative intensity, RER, and energy expenditure during aerobic exercise (n = 9)

	ME	HE
$V_{O_{2peak}}$ (%)	50 ± 1	69 ± 2*
HR (beat/min)	107 ± 5	128 ± 3*
Maximal HR (%)	63 ± 3	76 ± 1*
RER	0.84 ± 0.01	0.91 ± 0.01*
Total EE (kJ)	1529 ± 139	2117 ± 136*
Fat oxidation (kJ)	761 ± 77	633 ± 108*
Fat oxidation (%)	52 ± 5	30 ± 4*

Values are means ± SE. EE indicates energy expenditure.
* Significantly different from ME ($P < .05$).

Table 3
Blood hormone and metabolite concentrations (n = 9) and TNFα and IL-6 (n = 6) concentration during aerobic exercise

		Rest	30 min	60 min
Epinephrine	ME	273.0 ± 61.7	357.9 ± 86.9	509.6 ± 146.7
(pmol/L)	HE	266.9 ± 36.9	606.6 ± 73.4*,†	1146.6 ± 269.8*,†
Norepinephrine	ME	2.3 ± 0.3	4.7 ± 0.5†	5.6 ± 0.5†
(nmol/L)	HE	2.7 ± 0.3	8.8 ± 1.0*,†	12.1 ± 1.5*,†
Insulin	ME	57.2 ± 7.0	43.9 ± 5.6†	45.0 ± 5.1†
(pmol/L)	HE	62.1 ± 7.7	43.1 ± 6.2†	40.0 ± 4.71
Growth hormone	ME	2.7 ± 1.7	5.7 ± 2.1†	5.8 ± 1.4†
(μg/L)	HE	3.5 ± 1.9	8.6 ± 2.7†	9.3 ± 1.8†
FFA	ME	0.8 ± 0.1	1.6 ± 0.1†	1.9 ± 0.2†
(mmol/L)	HE	0.6 ± 0.1	2.2 ± 0.5*,†	2.5 ± 0.4*,†
Glycerol	ME	1.2 ± 0.2	6.6 ± 1.2†	7.4 ± 1.3†
(mmol/L)	HE	1.1 ± 0.1	9.9 ± 2.1*,†	11.0 ± 2.0*,†
Triglycerides	ME	1.4 ± 0.3	1.0 ± 0.3	1.0 ± 0.3
(mmol/L)	HE	1.9 ± 0.6	1.4 ± 0.5	1.2 ± 0.5
Glucose	ME	5.6 ± 0.2	5.2 ± 0.3	5.1 ± 0.3
(mmol/L)	HE	5.4 ± 0.1	5.0 ± 0.2	5.1 ± 0.2
TNFα	ME	9.3 ± 8.6	4.3 ± 3.1	3.0 ± 1.9
(pg/mL)	HE	4.2 ± 1.2	3.3 ± 1.2	3.0 ± 1.1
IL-6	ME	5.4 ± 4.1	1.7 ± 0.4	3.0 ± 1.1
(pg/mL)	HE	1.6 ± 0.9	1.6 ± 0.4	2.8 ± 0.5
Plasma volume	ME		0.28 ± 0.08	−0.66 ± 0.12
change (%)	HE		0.09 ± 0.11	−0.50 ± 0.10

Values are means ± SE.
* Significantly different from ME at the time ($P < .05$).
† Significantly different from rest at the same intensity ($P < .05$).

3.3. Blood metabolite responses

There were significant trial × time interactions for changes in serum FFA and glycerol concentrations ($P < .05$) (Table 3). The serum FFA and glycerol concentrations increased during both trials ($P < .05$), but were higher after HE than after ME ($P < .05$). There was no significant trial × time interaction for changes in the plasma glucose concentration. The serum triglyceride concentration tended to decrease with both ME and HE, but no significant trial × time interaction was evident.

3.4. Adiponectin responses

Serum total, HMW, and MLMW adiponectin concentration responses are shown in Figs. 1 and 2. Significant trial × time interactions were observed for changes in serum total and MLMW adiponectin concentrations ($P < .05$), but not for

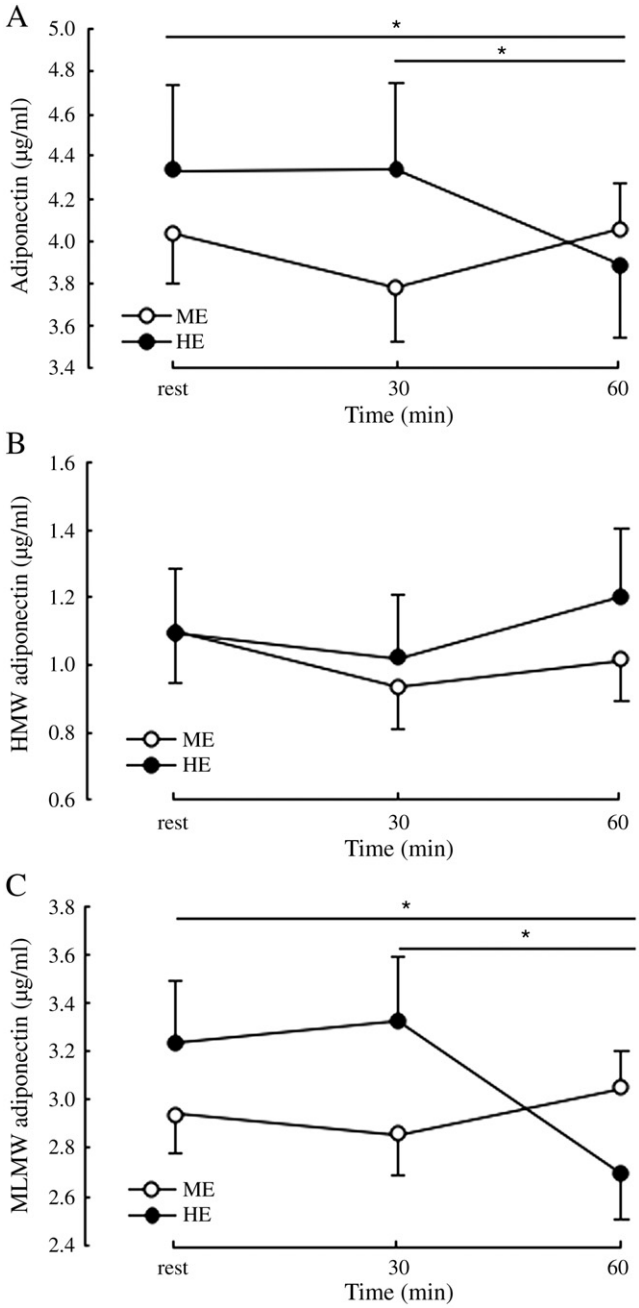

Fig. 1. Changes in total (A), HMW) (B), and MLMW (C) adiponectin concentrations during ME and HE. There was a significant trial × time interaction for the total and MLMW adiponectin concentrations ($P < .05$). No significant trial × time interactions were found for the HMW adiponectin concentration ($P > .05$). *Significantly different between rest and 60 minutes in HE ($P < .05$). Values are means ± SE.

the serum HMW adiponectin concentration. The total and MLMW adiponectin concentrations were lower at 60 minutes than during rest and at 30 minutes during HE ($P < .05$), but did not change during ME. The HMW adiponectin concentration remained unchanged during ME and HE (Fig. 1). The ratio of HMW to total adiponectin concentration increased and the ratio of MLMW to total adiponectin concentration decreased significantly during HE (Fig. 2).

3.5. Cytokines (IL-6 and TNFα) responses

IL-6 and TNFα results for 3 participants were excluded because of measurement errors. No significant trial × time interactions were found for changes in serum IL-6 and TNFα concentrations, and there were no changes in serum IL-6 or TNFα concentrations during ME and HE (Table 3).

3.6. Association between adiponectin and hormones, metabolites, and cytokines

During HE, the percentage changes of the total adiponectin and MLMW adiponectin concentrations were negatively correlated with the percentage changes in epinephrine concentration from rest to 60 min ($r = -0.67$, $P < .05$ and $r = -0.81$, $P < .05$, respectively; Fig. 3). The percentage changes of the total adiponectin and MLMW adiponectin concentrations were not correlated with the percentage changes of other hormones or cytokines from rest to 30 minutes or from

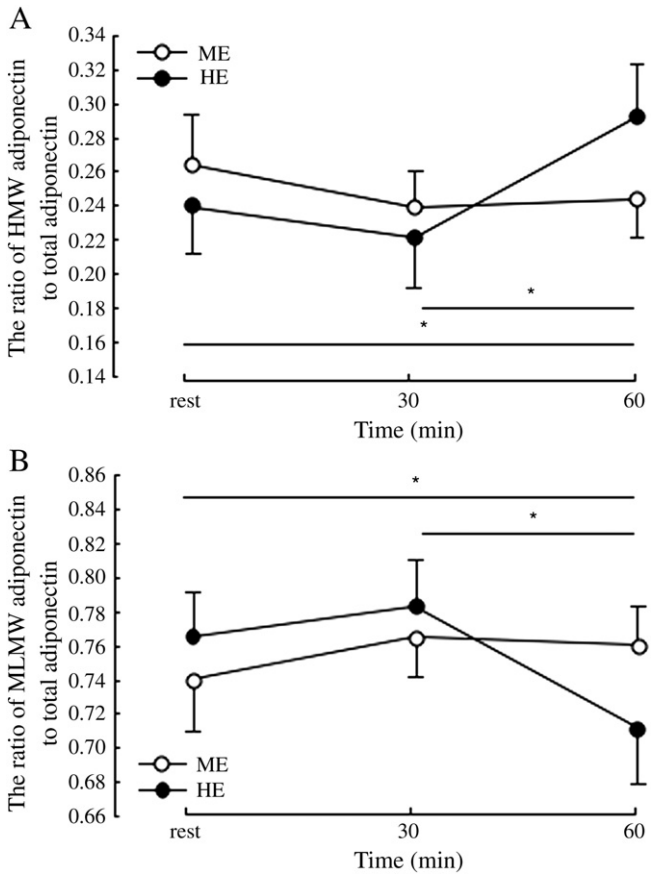

Fig. 2. Changes in the ratio of HMW (A) and MLMW (B) to total adiponectin concentration after ME and HE. Significant trial × time interactions were found for the ratios of HMW and MLMW to total adiponectin concentration ($P > .05$). *Significantly different between rest and 60 minutes in HE ($P < .05$). Values are means ± SE.

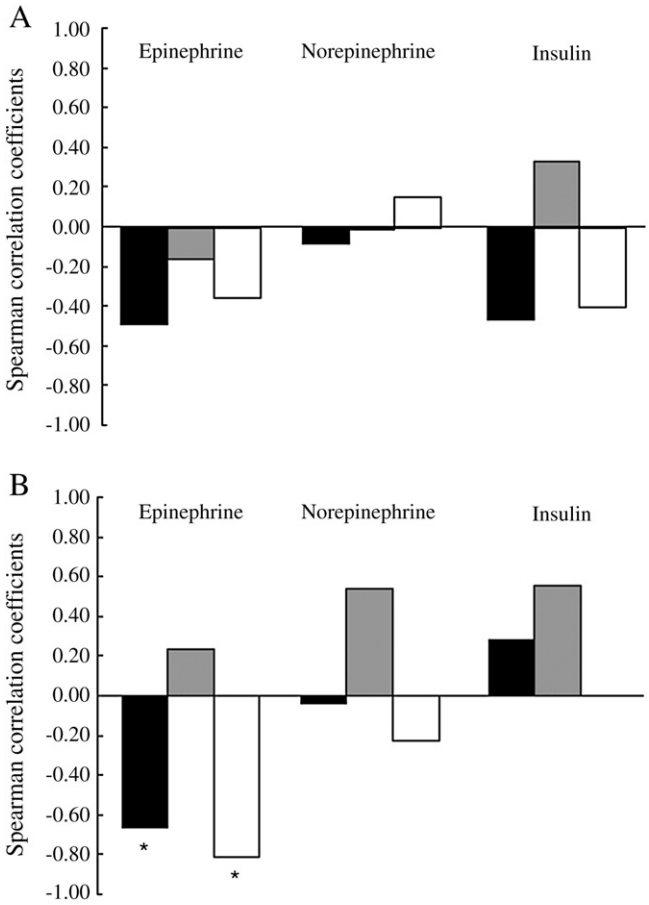

Fig. 3. Spearman correlation coefficients between changes in epinephrine, norepinephrine, and insulin concentration and change in total (black bar), HMW (gray bar), and MLMW (white bar) concentrations (A, 60-minute value − 30-minute value; B, 60-minute value − rest value). *$P < .05$. Values are means ± SE.

rest to 60 minutes. During ME, the percentage changes in total adiponectin or oligomer concentrations were not correlated with the percentage changes of any hormones, metabolites, or cytokines.

4. Discussion

The main finding of this study was that the total adiponectin concentration decreased during acute high-intensity aerobic exercise, but not during moderate-intensity aerobic exercise, in middle-aged abdominally obese men. The changes in total adiponectin concentration during high-intensity aerobic exercise were mainly due to changes in the MLMW adiponectin concentration. Differences in the responses of total adiponectin and MLMW adiponectin concentrations between exercise intensities may be due to differences in epinephrine concentration.

Our findings are in agreement with some previous studies [14,15], but contradict studies that reported unchanged plasma or serum concentrations of total adiponectin during acute aerobic exercise at intensities (50% Vo_{2max} to exhaustion) in healthy young men [16-21] and overweight young men [20]. However, the reason for these discrepancies remains unclear. Differences in age, fitness levels, and obesity phenotype may explain the contrasting results because these factors influence metabolism during exercise [39-42].

Adiponectin oligomer profiles do not change during moderate- to high-intensity aerobic exercise in healthy young men [19,21]. The present study is the first to investigate changes in adiponectin oligomers during moderate- and high-intensity aerobic exercise in middle-aged abdominally obese men. Several studies have demonstrated that HMW adiponectin is the more active form in liver and skeletal muscles compared with MMW and LMW adiponectin [6-8], suggesting that HMW adiponectin improves glucose and lipid metabolism after exercise; however, acute aerobic exercise did not affect the concentration of HMW adiponectin in the present study. This finding is in agreement with our previous study [21] and another study involving young men [19]. Accordingly, our data suggest that acute aerobic exercise does not affect the production or secretion of HMW adiponectin, and that the blood HMW adiponectin concentration may not affect glucose or lipid metabolism during acute exercise in middle-aged abdominally obese men.

The contribution of a decrease in total adiponectin accompanied by a selective decrease in MLMW adiponectin to metabolic disorders in obesity is currently not clear because LMW and MMW adiponectin has relatively weak effects on the liver and skeletal muscles [6-8]. However, O'Leary et al [43] recently found that the ratio of HMW to total adiponectin increased, with no change in absolute HMW adiponectin concentration, during 12 weeks of supervised aerobic exercise (60 minutes at 80%-85% maximum HR) without a reduction in caloric intake. The observed increase in the ratio was due to a reduction in the percentage change of the MMW adiponectin concentration. The authors also observed that the glucose disposal rate improved during the intervention. They proposed that a reduction in MMW adiponectin may lead to reduced competition between HMW and LMW adiponectin for binding to the adiponectin receptor. Their findings suggest that fluctuations in the levels of MMW adiponectin oligomers indirectly reinforce the function of HMW adiponectin and lead to improvements in metabolic disorders. In addition, this observation seems to be supported by our finding that the ratio of HMW to total adiponectin increased accompanied by decreasing concentrations of MLMW adiponectin during high-intensity aerobic exercise in an obese population.

The synthesis and secretion of adiponectin are partly controlled by several hormones, including insulin and catecholamines. Insulin may decrease adiponectin messenger RNA expression and protein secretion [44-46]. In

humans, the concentration of adiponectin decreases during hyperinsulinemic-euglycemic clamps [45,46]. However, in this study, the serum insulin concentration was similar with both exercise intensities; and the change in insulin concentration was not correlated with changes in adiponectin concentration. Therefore, the changes in total and MLMW adiponectin cannot be due to changes in insulin concentrations during HE. In contrast, the plasma concentrations of epinephrine and norepinephrine were higher during HE than during ME. Furthermore, the changes in epinephrine concentration were negatively correlated with the total and MLMW adiponectin concentrations, suggesting that epinephrine inhibits the release of total and MLMW adiponectin during HE. Previous studies have reported that increased β-adrenergic activation down-regulates adiponectin messenger RNA, protein synthesis, and secretion in cell lines [44], in animals [25-28], and in human adipose tissue in vitro [25] and that the down-regulation of adiponectin secretion may be induced by an increase in cyclic AMP as a second messenger in the lipolytic cascade in adipocytes [25]. Therefore, it is possible that the marked increase in cyclic AMP in adipose tissue due to an increase in circulating catecholamine concentration during HE attenuates MMW or LMW adiponectin secretion from adipocytes.

Proinflammatory cytokines also affect adiponectin levels. IL-6 [30] and TNFα [29] interfere with adiponectin production and secretion. In particular, IL-6 increases during acute exercise [17,20,32] in proportion to exercise intensity [31]. In the present study, the concentrations of IL-6 and TNFα remained unchanged and were similar in both exercise intensities. Therefore, it seems unlikely that IL-6 and TNFα can account for the differences in adiponectin oligomer concentrations between the 2 exercise intensities.

There are several limitations to this study. First, this study was not a controlled trial; and the influence of circadian rhythms cannot be entirely excluded. The circulating concentration of adiponectin shows ultradian pulsatility and diurnal variation [47-49], although the adiponectin concentration remains stable during daytime (8:00 AM to 8:00 PM). Similarly, the plasma concentration of catecholamines also shows circadian rhythms [50,51]. In particular, epinephrine reaches a peak level in the morning at around 8:00 AM [50]; and the relative change from the peak to the trough is 53% [51]. The change in epinephrine concentration during ME was within this range, whereas the change during HE clearly exceeds this range. Therefore, the observed changes in adiponectin and epinephrine concentration cannot be explained only by the ultradian pulsatility or diurnal variation in adiponectin or epinephrine. Nevertheless, studies based on a controlled trial design are required to further investigate our findings. Second, the participants in our study were sedentary, middle-aged, abdominally obese men. Our findings do not apply to other individuals such as healthy young or old men and women, obese women, or populations with abnormal metabolism. Finally, the total adiponectin and adiponectin oligomer concentrations were measured at 3 points during aerobic exercise. If sampling was performed more frequently, the continuous time course of changes in adiponectin, hormones, cytokines, and metabolites during aerobic exercise could be determined. In addition, the decreased adiponectin concentration might recover above baseline levels after exercise [14]; consequently, it is unclear whether the concentration of adiponectin oligomers changed after exercise in our study. Measurement of blood adiponectin concentrations after exercise would provide greater insight into the responses of adiponectin to exercise.

In conclusion, the results of our study demonstrate that high-intensity aerobic exercise altered total and MLMW adiponectin concentrations in middle-aged abdominally obese men. These findings suggest that the MLMW adiponectin concentration, rather than the HMW adiponectin concentration, is influenced by changes in epinephrine concentration during exercise in middle-aged abdominally obese men. Additional studies are needed to determine whether changes in adiponectin oligomer concentrations influence metabolism after acute exercise and chronic exercise training.

Acknowledgment

This research was supported in part by a Grant-in-Aid for Young Scientists Start-up (19800047) awarded by the Japan Society for the Promotion of Science of Japan; the 21st Century Center of Excellence program (2002–2003 Nishihira/Tsukuba Project: Promotion of Health and Sport Scientific Research) run by the Ministry of Education, Culture, Sports, Science, and Technology of Japan; and the 2004-2006 Tanaka Project of the Tsukuba Advanced Research Alliance, University of Tsukuba.

References

[1] Katagiri H, Yamada T, Oka Y. Adiposity and cardiovascular disorders: disturbance of the regulatory system consisting of humoral and neuronal signals. Circ Res 2007;101:27-39.

[2] Matsushita K, Yatsuya H, Tamakoshi K, et al. Comparison of circulating adiponectin and proinflammatory markers regarding their association with metabolic syndrome in Japanese men. Arterioscler Thromb Vasc Biol 2006;26:871-6.

[3] Pischon T, Girman CJ, Hotamisligil GS, et al. Plasma adiponectin levels and risk of myocardial infarction in men. JAMA 2004;291: 1730-7.

[4] Yamauchi T, Kamon J, Minokoshi Y, et al. Adiponectin stimulates glucose utilization and fatty-acid oxidation by activating AMP-activated protein kinase. Nat Med 2002;8:1288-95.

[5] Yamamoto Y, Hirose H, Saito I, et al. Adiponectin, an adipocyte-derived protein, predicts future insulin resistance: two-year follow-up study in Japanese population. J Clin Endocrinol Metab 2004;89: 87-90.

[6] Pajvani UB, Du X, Combs TP, et al. Structure-function studies of the adipocyte-secreted hormone Acrp30/adiponectin. Implications for metabolic regulation and bioactivity. J Biol Chem 2003;278: 9073-85.

[7] Waki H, Yamauchi T, Kamon J, et al. Impaired multimerization of human adiponectin mutants associated with diabetes. Molecular structure and multimer formation of adiponectin. J Biol Chem 2003; 278:40352-63.
[8] Hada Y, Yamauchi T, Waki H, et al. Selective purification and characterization of adiponectin multimer species from human plasma. Biochem Biophys Res Commun 2007;356:487-93.
[9] Hara K, Horikoshi M, Yamauchi T, et al. Measurement of the high–molecular weight form of adiponectin in plasma is useful for the prediction of insulin resistance and metabolic syndrome. Diabetes Care 2006;29:1357-62.
[10] Lara-Castro C, Luo N, Wallace P, et al. Adiponectin multimeric complexes and the metabolic syndrome trait cluster. Diabetes 2006;55: 249-59.
[11] Seino Y, Hirose H, Saito I, Itoh H. High–molecular-weight adiponectin is a predictor of progression to metabolic syndrome: a population-based 6-year follow-up study in Japanese men. Metabolism 2009;58:355-60.
[12] von Eynatten M, Humpert PM, Bluemm A, et al. High–molecular weight adiponectin is independently associated with the extent of coronary artery disease in men. Atherosclerosis 2008;199:123-8.
[13] American College of Sports Medicine. Health appraisal, risk assessment, and safety of exercise. ACSM's guidelines for exercise testing and prescription, 7th edPhiladelphia: Lippincott Williams & Wilkins; 2006. p. 3-35.
[14] Jürimäe J, Purge P, Jürimäe T. Adiponectin is altered after maximal exercise in highly trained male rowers. Eur J Appl Physiol 2005;93: 502-5.
[15] Jürimäe J, Purge P, Jürimäe T. Adiponectin and stress hormone responses to maximal sculling after volume-extended training season in elite rowers. Metabolism 2006;55:13-9.
[16] Kraemer RR, Aboudehen KS, Carruth AK, et al. Adiponectin responses to continuous and progressively intense intermittent exercise. Med Sci Sports Exerc 2003;35:1320-5.
[17] Ferguson MA, White LJ, McCoy S, et al. Plasma adiponectin response to acute exercise in healthy subjects. Eur J Appl Physiol 2004;91: 324-9.
[18] Punyadeera C, Zorenc AH, Koopman R, et al. The effects of exercise and adipose tissue lipolysis on plasma adiponectin concentration and adiponectin receptor expression in human skeletal muscle. Eur J Endocrinol 2005;152:427-36.
[19] Bobbert T, Wegewitz U, Brechtel L, et al. Adiponectin oligomers in human serum during acute and chronic exercise: relation to lipid metabolism and insulin sensitivity. Int J Sports Med 2007;28: 1-8.
[20] Højbjerre L, Rosenzweig M, Dela F, et al. Acute exercise increases adipose tissue interstitial adiponectin concentration in healthy overweight and lean subjects. Eur J Endocrinol 2007;157:613-23.
[21] Numao S, Suzuki M, Matsuo T, et al. Effects of acute aerobic exercise on high-molecular-weight adiponectin. Med Sci Sports Exerc 2008;40: 1271-6.
[22] Romijn JA, Coyle EF, Sidossis LS, et al. Regulation of endogenous fat and carbohydrate metabolism in relation to exercise intensity and duration. Am J Physiol 1993;265:E380-91.
[23] Kanaley JA, Mottram CD, Scanlon PD, Jensen MD. Fatty acid kinetic responses to running above or below lactate threshold. J Appl Physiol 1995;79:439-47.
[24] Fasshauer M, Klein J, Neumann S, et al. Adiponectin gene expression is inhibited by beta-adrenergic stimulation via protein kinase A in 3T3-L1 adipocytes. FEBS Lett 2001;507:142-6.
[25] Delporte ML, Funahashi T, Takahashi M, et al. Pre- and post-translational negative effect of beta-adrenoceptor agonists on adiponectin secretion: in vitro and in vivo studies. Biochem J 2002;367: 677-85.
[26] Imai J, Katagiri H, Yamada T, et al. Cold exposure suppresses serum adiponectin levels through sympathetic nerve activation in mice. Obesity 2006;14:1132-41.
[27] Cong L, Chen K, Li J, et al. Regulation of adiponectin and leptin secretion and expression by insulin through a PI3K-PDE3B dependent mechanism in rat primary adipocytes. Biochem J 2007; 403:519-25.
[28] Fu L, Isobe K, Zeng Q, et al. Beta-adrenoceptor agonists downregulate adiponectin, but upregulate adiponectin receptor 2 and tumor necrosis factor-alpha expression in adipocytes. Eur J Pharmacol 2007;569: 155-62.
[29] Fasshauer M, Kralisch S, Klier M, et al. Adiponectin gene expression and secretion is inhibited by interleukin-6 in 3T3-L1 adipocytes. Biochem Biophys Res Commun 2003;301:1045-50.
[30] Bruun JM, Lihn AS, Verdich C, et al. Regulation of adiponectin by adipose tissue-derived cytokines: in vivo and in vitro investigations in humans. Am J Physiol Endocrinol Metab 2003;285:E527-33.
[31] Helge JW, Stallknecht B, Pedersen BK, et al. The effect of graded exercise on IL-6 release and glucose uptake in human skeletal muscle. J Physiol 2003;546:299-305.
[32] Petersen AM, Pedersen BK. The anti-inflammatory effect of exercise. J Appl Physiol 2005;98:1154-62.
[33] Examination Committee of Criteria for 'Obesity Disease' in Japan, Japan Society for the Study of Obesity. New criteria for 'obesity disease' in Japan. Circ J 2002;66:987-92.
[34] Tanaka K, Takeshima N, Kato T, et al. Critical determinants of endurance performance in middle-aged and elderly endurance runners with heterogeneous training habits. Eur J Appl Physiol Occup Physiol 1990;59:443-9.
[35] Nakata Y, Ohkawara K, Lee DJ, et al. Effects of additional resistance training during diet-induced weight loss on bone mineral density in overweight premenopausal women. J Bone Miner Metab 2008;26: 172-7.
[36] Peronnet F, Massicotte D. Table of nonprotein respiratory quotient: an update. Can J Sport Sci 1991;16:23-9.
[37] Dill DB, Costill DL. Calculation of percentage changes in volumes of blood, plasma and red cells in dehydration. J Appl Physiol 1974;37: 247-8.
[38] Ebinuma H, Miyazaki O, Yago H, et al. A novel ELISA system for selective measurement of human adiponectin multimers by using proteases. Clin Chim Acta 2006;372:47-53.
[39] Kanaley JA, Cryer PE, Jensen MD. Fatty acid kinetic responses to exercise. Effects of obesity, body fat distribution, and energy-restricted diet. J Clin Invest 1993;92:255-61.
[40] Klein S, Coyle EF, Wolfe RR. Fat metabolism during low-intensity exercise in endurance-trained and untrained men. Am J Physiol 1994; 267:E934-40.
[41] Toth MJ, Arciero PJ, Gardner AW, et al. Rates of free fatty acid appearance and fat oxidation in healthy younger and older men. J Appl Physiol 1996;80:506-11.
[42] Numao S, Hayashi Y, Katayama Y, et al. Effects of obesity phenotype on fat metabolism in obese men during endurance exercise. Int J Obes 2006;30:1189-96.
[43] O'Leary VB, Jorett AE, Marchetti CM, et al. Enhanced adiponectin multimer ratio and skeletal muscle adiponectin receptor expression following exercise training and diet in older insulin-resistant adults. Am J Physiol Endocrinol Metab 2007;293:E421-7.
[44] Fasshauer M, Klein J, Neumann S, et al. Hormonal regulation of adiponectin gene expression in 3T3-L1 adipocytes. Biochem Biophys Res Commun 2002;290:1084-9.
[45] Möhlig M, Wegewitz U, Osterhoff M, et al. Insulin decreases human adiponectin plasma levels. Horm Metab Res 2002;34:655-8.
[46] Blümer RM, van der Crabben SN, Stegenga ME, et al. Hyperglycemia prevents the suppressive effect of hyperinsulinemia on plasma adiponectin levels in healthy humans. Am J Physiol Endocrinol Metab 2008;295:E613-7.
[47] Gavrila A, Peng CK, Chan JL, et al. Diurnal and ultradian dynamics of serum adiponectin in healthy men: comparison with leptin, circulating soluble leptin receptor, and cortisol patterns. J Clin Endocrinol Metab 2003;88:2838-43.

[48] Yildiz BO, Suchard MA, Wong ML, et al. Alterations in the dynamics of circulating ghrelin, adiponectin, and leptin in human obesity. Proc Natl Acad Sci U S A 2004;101:10434-9.

[49] Shea SA, Hilton MF, Orlova C, et al. Independent circadian and sleep/wake regulation of adipokines and glucose in humans. J Clin Endocrinol Metab 2005;90:2537-44.

[50] Dimitrov S, Benedict C, Heutling D, et al. Cortisol and epinephrine control opposing circadian rhythms in T cell subsets. Blood 2009;113:5134-43.

[51] Scheer FA, Hilton MF, Mantzoros CS, Shea SA. Adverse metabolic and cardiovascular consequences of circadian misalignment. Proc Natl Acad Sci U S A 2009;106:4453-8.