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Visceral and Subcutaneous Adiposity and Adiponectin in Middle-aged Japanese Men: The ERA JUMP Study

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Abstract

Adiponectin is reduced in obesity, and has been suggested to play an important role in modulation of atherosclerosis. We studied the relationship between visceral (VAT) and subcutaneous (SAT) adipose tissue and serum adiponectin concentrations in Japanese men. Participants were 304 randomly selected community-based Japanese men aged 40 to 49 without a prior history of cardiovascular disease. Participants were grouped according to tertiles of serum adiponectin. In multiple linear regression analysis including age, pack years of smoking, and alcohol intake as covariates, log-transformed adiponectin was **inversely** associated with both VAT and SAT when these two obesity measures were included separately in the models. However, log-transformed adiponectin was **inversely** associated with VAT (standardized β estimate= −0.465, P< 0.0001) and **positively** associated with SAT (standardized β estimate = 0.166, P=0.03), when these were included concomitantly in the model. In conclusion, VAT and SAT had differential associations with serum adiponectin concentrations.

INTRODUCTION

An adipose-specific protein, adiponectin, is reduced in obesity, insulin resistance and type 2 diabetes; plasma concentrations are inversely related to body weight, especially visceral adiposity [1-4]. Adiponectin has been suggested to play an important role in modulating atherosclerosis [5-7].

Although body mass index (BMI) is an indicator of overall adiposity, different fat compartments have been proposed to be associated with differential metabolic risk [8]. Recently, evidence indicating visceral adipose tissue (VAT) is more strongly associated with an adverse metabolic risk profile than subcutaneous adipose tissue (SAT) is accumulating [9-13].

We studied the relationship between VAT and SAT and serum adiponectin concentrations in community-based middle-aged Japanese men. On the basis of previous studies [9-13], we

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hypothesized that the association of VAT with serum adiponectin concentration would be different from that of SAT.

METHODS

Participants

Participants were randomly selected men aged 40-49 from Kusatsu City, Shiga Prefecture, Japan, using information from the Basic Residents' Register. Exclusion criteria included: 1) clinical cardiovascular disease, 2) type 1 diabetes or other severe diseases [14]. From May 2001 to February 2004, randomly selected men from the Register were contacted consecutively via phone. The rate of participation was about 49%. Informed consent was obtained from all 304 eligible participants [14]. The Institutional Review Board of Shiga University Medical Science approved this study.

Baseline Examinations and Lifestyle Assessments

Blood pressure was measured in the right arm of the seated participants after they were seated quietly for five minutes using an automated sphygmomanometer (BP-8800, Colin Medical Technology, Komaki, Japan). The average of two measurements was used. Body weight and height were measured while the participants were wearing light clothing and in stocking feet. Waist circumference (WC) was measured with a tape measure at the level of the umbilicus while the participants were standing and at the end of exhalation.

A lifestyle survey was carried out using a self-administered questionnaire. Current smoking was defined as smoking cigarettes over the last month. Pack-years were calculated as years of smoking multiplied by the number of cigarettes per day divided by 20. Alcohol drinkers were defined as drinking alcohol two days per week or more. Ethanol consumption per day was estimated assuming that concentrations of alcohol were 5% for beer, 12% for wine, 40% for liquor, 16% for sake (Japanese rice wine) and 25% for shochu (Japanese spirits made from barley, sweet potato, or rice or any combination of these) [16].

Biochemical measurements

Venipuncture was performed early in the clinic visit after a 12-hour fast. Plasma or serum samples were prepared and frozen at –80 °C. The samples were shipped on dry ice to the Heinz Laboratory, University of Pittsburgh. Serum low-density-lipoprotein cholesterol (LDLc), high-density-lipoprotein cholesterol (HDLc), triglycerides, glucose and C-reactive protein (CRP) were measured as previously described [15]. Serum adiponectin was measured with a radioimmunoassay procedure (Linco Research, Inc, St Charles, MO). An insulin resistance index, the homeostasis model assessment of insulin resistance (HOMA-R), was obtained using insulin (μU/ml) × fasting blood glucose (mg/dl)/405 [17].

Abdominal Adipose Tissue Measurements

Electron-beam tomography (EBCT) scanning was done using a GE Imatron C150 scanner (GE Medical System, South San Francisco, CA). Each participant was positioned supine on a table with the head toward the gantry and a bolster pillow was placed under the knees for comfort. The scanner was set in continuous volume scanning (CVS) mode so that gating was not required. For VAT and SAT, a slice between L4/L5 was used. Scan data were saved to optical disc. All EBCT studies were read centrally at the University of Pittsburgh. The VAT area was determined on an Imatron workstation using software obtained from Accuimage Diagnostic Corporation. Using a pixel range of −190 to −30 Housfield Unit (HU) as the range for fat, the area of adipose tissue within the region of interest was determined using image analysis. The fat within this circle was considered to be VAT. The fat area for the entire image was then

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determined and the difference in fat area between the whole image and VAT was equal to the SAT. The intraclass correlation coefficients were 0.99 for SAT and 0.99 for VAT.

Statistical Analysis

SAS version 9.1 for Windows (SAS Institute, Cary, NC) was used. Because the distributions of serum adiponectin, triglycerides and CRP were positively skewed, a logarithmic transformation was used to normalize the distribution. The Mantel-Haenszel chi-square statistical test was used to detect deviation from linearity in the association between nominal variables and the three groups according to tertiles of serum adiponectin concentration. The "contrast" option for analysis of variance was used to detect deviation from linearity in the association between continuous variables and the three groups.

Spearman's partial correlation coefficients were calculated for log-transformed adiponectin, BMI, WC, VAT, SAT and log-transformed CRP, HDLc, LDLc, log-transformed triglycerides, HOMA-R, smoking (pack-years of smoking) and alcohol consumption per day, after adjustment for age.

Multiple linear regression analysis was used to examine the relationship between log-transformed adiponectin and VAT or SAT adjusting for confounders. Basic covariates used for adjustment were age, smoking (pack year) and alcohol (g/day). Model 1 included basic covariates + VAT; Model 2, basic covariates + SAT; Model 3, basic covariates + VAT,SAT; Model 4, basic covariates + VAT, BMI; Model 5, basic covariates + VAT, WC; Model 6, basic covariates + VAT, BMI, WC; Model 7, basic covariates + SAT, BMI; Model 8, basic covariates + SAT, WC; Model 9, basic covariates + SAT, BMI, WC. Similar analyses were performed with log-transformed CRP and HOMA-R in associations with VAT and SAT.

All P values were two-tailed and $P<0.05$ was considered significant. Data are presented as the mean ± standard deviation (SD) unless stated otherwise.

RESULTS

Characteristics of Participants According to Tertiles of Serum Adiponectin Concentration

Characteristics of participants according to tertiles of serum adiponectin concentration are shown in Table 1. The mean age, systolic blood pressure, diastolic blood pressure, current smoker status, alcohol intake and LDLc were not statistically different among the groups. As expected, the four obesity measures (BMI, WC, VAT, and SAT) decreased in the higher adiponectin concentration groups. The mean HDLc increased in the higher adiponectin concentration groups. The mean triglycerides, CRP (both log-transformed in the analysis) and HOMA-R fell significantly in the higher adiponectin concentration groups.

Correlation Between the Obesity Measures and Log-Transformed Adiopnectin, and Other Variables

All four obesity measures, BMI, WC, VAT and SAT were highly correlated with each other (R: 0.700 to 0.899, all $P<0.0001$). All four obesity measures were significantly positively correlated with log-transformed CRP, log-transformed triglycerides, and HOMA-R and significantly inversely correlated with log-transformed adiponectin and HDLc. Except for VAT, the three other obesity measures were significantly positively correlated with LDLc (Table 2). Except for asignificant positive correlation between VAT and alcohol intake, smoking and alcohol intake were not consistently correlated with the obesity measures.

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Multivariate-Adjusted Linear Regression Models of the Relationship of VAT or SAT to Log-Transformed Adiopnectin (Table 3)

In multivariate models, log-transformed adiponectin was significantly inversely associated with both VAT and SAT when these two obesity measures were included separately in the models (Model 1 and 2). However, when these were included together in the model, log-transformed adiponectin was significantly inversely associated with VAT, but significantly **positively** associated with SAT (Model 3). Differential associations of VAT and SAT with log-transformed adiponectin were also seen when these two obesity measures were included separately, but concomitantly with BMI or WC in some models (Model 4 through 8) and concomitantly with both BMI and WC in another model (Model 9). Furthermore, in Model 7, the statistical significance of the SAT association with log-transformed adiponectin was lost.

Similar analyses were performed with log-transformed CRP and HOMA-R in association with VAT and SAT. Both variables were significantly positively associated with VAT and SAT. However, no differential associations as seen for log-transformed adiponectin were noted with these two variables (data not shown).

DISCUSSION

Our study in a population-based sample of Japanese middle-aged men showed differential associations of VAT and SAT with adiponectin when VAT and SAT were included concomitantly in a regression model; VAT had a significant inverse and SAT a significant positive, association. The differential associations were also seen when these two obesity measures were included separately, but concomitantly, with BMI and WC in the models. However, VAT or SAT had a significant inverse association with adiponectin when each was entered into the model without BMI or WC. Previous studies on the associations of VAT and SAT with inflammation markers or metabolic risk factors found significant associations of these two obesity measures with those variables [11,12]. Concomitant inclusion of the two obesity measures, or a combination with BMI and WC, showed either of these two remained significant or one of the two became insignificant. No previous study has reported any differential associations with these variables. Pou et al. studied the relations of VAT and SAT with circulating inflammatory and oxidative stress biomarkers in Framingham Heart Study participants and found that SAT ad VAT were positively and similarly related to CRP, fibrinogen, intercellular adhesion molecule-1, interleukin, P-selectin and tumor necrosis factor receptor-2. On the other hand, VAT was more strongly associated with urinary isoprostanes and monocyte chemoattractant protein-1 than with SAT. When BMI and WC were added to the models, VAT remained significantly associated with only CRP, interleukin-6, isoprostanes and monocytes chemoattractant protein-1; SAT only remained associated with fibrinogen [13]. Fox et al. studied the association of VAT and SAT with metabolic risk factors in Framingham Heart Study participants and found that VAT was more strongly correlated with most metabolic factors than SAT [12].

Because all four obesity measures were highly correlated with each other, it is understandable that each had inverse associations with serum adiponectin concentrations when analyzed separately. Differential associations of VAT and SAT with adiponectin were found when these were included concomitantly in the model, indicating SAT may be positively associated with serum adiponectin independently. In a previous study on the relationship between serum adiponectin and leptin concentrations and body fat distribution, it was found that adiponectin was more strongly influenced by VAT, whereas leptin was more strongly influenced by SAT [18]. The differential associations of VAT and SAT with adiponectin were not examined. It has been considered that VAT is a pathogenic adipose tissue compartment rather than SAT. One of the mechanisms for this is thought to be that sustained exposure of the liver to an increased flux of free fatty acid via the portal circulation from VAT is antecedent to

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disturbances in glucose and lipid metabolism [19,20]. The other mechanism is related to the fact that most adipocytokines are secreted to a different extent in different adipose tissue depots, and for the most part, VAT is a more active producer of adipocytokines than SAT [21-23]. However, more leptin is secreted by SAT than VAT [24,25].

Body fat distribution differs across ethnic groups and some studies suggested Asians had more VAT than expected for a given BMI [26]. In a study by Lear et al., the mean BMI, VAT and SAT for European and Chinese participants were 27.7 kg/m^2, 100.8 cm^2, 265.9 cm^2, and 25.7 kg/m^2, 100.0 cm^2, 221.2 cm^2, respectively [27].

Strengths and Limitations

The main strengths of the present study were: (1) a population-based random sample; and (2) the age of our participants, 40-49 years, was narrow, so we could minimize the confounding effects generated from participant age. The study was limited by (1) its cross-sectional design, thereby precluding any inference of a causal relation between VAT, SAT and serum adiponectin concentrations; (2) the results may not be easy to generalize to other sex, ethnic or age groups because our sample was limited to middle-aged Japanese men living in Japan; (3) a relatively small sample size with a participation rate of about 50%. For a study that includes relatively new markers of continuous variables, adiponectin, VAT and SAT, the sample size appears adequate. The mean BMI in the present study was 23.7 kg/m^2, which was comparable with that of previous studies with a larger number of Japanese participants [28, 29]. The participation rate in the present study was about 50%, which is also comparable to current epidemiological studies [30,31]. For instance, the participation rate of the Multi-Ethnic Study of Atherosclerosis (MESA) was reported to be about 30% [30].

Conclusion

VAT and SAT had differential associations with serum adiponectin concentrations. This may be related to some of the mechanisms for VAT's stronger association with an adverse metabolic risk profile than SAT.

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Table 1

Characteristics of the Participants According to Tertiles of Serum Adiponectin Concentration, 304 Japanese men, 2001-04.

	Tertile 1	Tertile 2	Tertile 3	Trend P
Adiponectin range (μg/ml)	1.625-4.945	4.95-7.73	7.76-40.94	
Number	101	102	101	
Age (years)	44.7±2.7	45.4±2.7	45.2±3.0	0.269
SBP (mmHg)	124±13	127±18.6	124±16	0.858
DBP (mmHg)	76±11	78±13	75±11	0.364
BMI (kg/m^2)	24.7±2.8	23.8±3.1	22.7±3.0	<0.0001
WC (cm)	87.7±7.1	86.0±8.5	82.2±7.7	<0.0001
VAT (cm^2)	90.7±25.9	83.6±31.4	65.8±28.7	<0.0001
SAT (cm^2)	89.7±34.4	85.1±35.4	72.0±35.0	0.0004
Current smoker (%)	46.5	48.0	52.5	0.399
Smoking (pack years)	19.6±17.8	19.4±17.2	20.3±16.3	0.763
Alcohol intake (g/day)	24.7±27.2	29.0±30.1	26.3±28.6	0.701
LDLc (mg/dl)	132.7±33.0	134.1±33.9	130.4±41.3	0.772
HDLc (mg/dl)	49.0±13.4	53.8±11.0	59.5±14.4	<0.0001
Triglycerides* (mg/dl)	155 (127, 206)	138 (110, 184)	118 (87, 150)	<0.0001
HOMA-R	3.4±2.0	2.6±1.2	2.3±1.1	<0.0001
CRP* (mg/l)	0.44 (0.19, 0.83)	0.27 (0.15, 0.51)	0.26 (0.15, 0.53)	0.025

Values are mean±SD, percent, or median* (25^{th}, and 75^{th} percentile). SBP=systolic blood pressure, DBP= diastolic blood pressure, BMI=body mass index, WC=waist circumference, VAT= visceral adipose tissue area, SAT= subcutaneous adipose tissue area, LDLc=low-density lipoprotein cholesterol, HDLc= high-density lipoprotein cholesterol, HOMA-R= homeostasis model assessment insulin resistance index, CRP=C-reactive protein

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Table 2

Partial Correlation Coefficients Between the Obesity Measures and Log-Transformed Adiopnectin and Other Variables (Age-Adjusted), 304 Japanese men, 2001-04.

	logAdipo	BMI	WC	VAT	SAT
logAdipo	1.000	−0.237 (<0.0001)	−0.251 (<0.0001)	−0.335 (<0.0001)	−0.155 (0.007)
logCRP	−0.111 (0.055)	0.262 (<0.0001)	0.290 (<0.0001)	0.286 (<0.0001)	0.282 (<0.0001)
HDLc	0.292 (<0.0001)	−0.380 (<0.0001)	−0.397 (<0.0001)	−0.363 (<0.0001)	−0.354 (<0.0001)
LDLc	−0.006 (0.917)	0.137 (0.018)	0.147 (0.011)	0.078 (0.179)	0.188 (0.001)
logTG	−0.249 (<0.0001)	0.295 (<0.0001)	0.346 (<0.0001)	0.451 (<0.0001)	0.264 (<0.0001)
HOMA-R	−0.235 (<0.0001)	0.467 (<0.0001)	0.404 (<0.0001)	0.372 (<0.0001)	0.377 (<0.0001)
Smoking (pack years)	−0.032 (0.587)	−0.037 (0.525)	−0.015 (0.792)	−0.005 (0.938)	−0.062 (0.285)
Alcohol intake (g/day)	0.018 (0.762)	0.064 (0.269)	0.101 (0.080)	0.148 (0.010)	0.017 (0.770)

Partial correlation coefficients and (P values). logAdipo=log-transformed adiponectin concentration, BMI=body mass index, WC=waist circumference, VAT= visceral adipose tissue area, SAT= subcutaneous adipose tissue area, HDLc= high-density lipoprotein cholesterol, LDLc=low-density lipoprotein cholesterol, TG=triglycerides, HOMA-R= homeostasis model assessment insulin resistance index, CRP=C-reactive protein

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Table 3

Multivariates-Adjusted Linear Regression Models of Relationship of VAT or SAT to Log-Transformed Adiopnectin, 304 Japanese men, 2001-04.

Model	Covariates	VAT Standardized β	P	SAT Standardized β	P
Model 1	Basic + VAT	–0.347	<0.0001	-	-
Model 2	Basic + SAT	-	-	–0.157	0.007
Model 3	Basic + VAT,SAT	–0.465	<0.0001	0.166	0.030
Model 4	Basic + VAT, BMI	–0.362	<0.0001	-	-
Model 5	Basic + VAT, WC	–0.376	<0.0001	-	-
Model 6	Basic + VAT, BMI, WC	–0.375	<0.0001	-	-
Model 7	Basic + SAT, BMI	-	-	0.168	0.117
Model 8	Basic + SAT, WC	-	-	0.366	0.004
Model 9	Basic + SAT, BMI, WC	-	-	0.397	0.002

Basic: basic covariates used for adjustment were age, smoking (pack year) and alcohol intake (g/day).

BMI=body mass index, WC=waist circumference, VAT= visceral adipose tissue area, SAT= subcutaneous adipose tissue area.

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