Regional Abdominal Fat Distribution in Lean and Obese Thai Type 2 Diabetic Women: Relationships With Insulin Sensitivity and Cardiovascular Risk Factors

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To determine the relationships of body fat distribution and insulin sensitivity and cardiovascular risk factors in lean and obese Thai type 2 diabetic women, 9 lean and 11 obese subjects, with respective mean age 41.7 ± 6.3 (SD) and 48.0 ± 8.5 years, and mean body mass index (BMI) 23.5 ± 1.8 and 30.3 ± 3.7 kg/m^2, were studied. The amount of total body fat (TBF) and total abdominal fat (AF) were measured by dual-energy x-ray absorptiometer, whereas subcutaneous (SAF) and visceral abdominal fat areas (VAF) were measured by computerized tomography (CT) of the abdomen at the L4-L5 level. Insulin sensitivity was determined by euglycemic hyperinsulinemic clamp. Cardiovascular risk factors, which included fasting and post-glucose challenged plasma glucose and insulin, systolic (SBP) and diastolic blood pressure (DBP), lipid profile, fibrinogen, and uric acid, were also determined. VAF was inversely correlated with insulin sensitivity as determined by glucose infusion rate (GIR) during the clamp, in both lean ($r = -0.8821$; $P = .009$) and obese subjects ($r = -0.582$; $P = .078$) independent of percent TBF. SAF and TBF were not correlated with GIR. With regards to cardiovascular risk factors, VAF was correlated with SBP ($r = 0.5279$; $P = .024$) and DBP ($r = 0.6492$; $P = .004$), fasting insulin ($r = 0.7256$; $P = .001$) and uric acid ($r = 0.4963$; $P = .036$) after adjustment for percent TBF. In contrast, TBF was correlated with fasting insulin ($r = 0.517$; $P = .023$), area under the curve (AUC) of insulin ($r = 0.625$; $P = .004$), triglyceride (TG) ($r = 0.668$; $P = .002$), and uric acid ($r = 0.49$; $P = .033$). GIR was not correlated with any of cardiovascular risk factors independent of VAF. In conclusion, VAF was a strong determinant of insulin sensitivity and several cardiovascular risk factors in both lean and obese Thai type 2 diabetic women.

THE ASSOCIATION of body fat, particularly abdominal fat (AF) and insulin sensitivity, as well as cardiovascular risk, has been recognized in both nondiabetic and diabetic subjects.[1-3] Several, but not all, studies, mostly in Caucasian populations, demonstrated stronger positive relationships between the amount of intra-abdominal or visceral abdominal fat (VAF) and insulin resistance and several cardiovascular risk factors than that of total body fat (TBF) in patients with type 2 diabetes, particularly obese patients.[4-6] However, since the level of VAF may differ among various populations, this relationship may not be observed or may be less strong in a non-Caucasian population.[7,8] Studies in Asian type 2 diabetes also demonstrated the strong inverse relationship of VAF and insulin sensitivity in obese patients.[9,11] In contrast, Taniguchi et al[12] reported a stronger inverse relationship of subcutaneous abdominal fat (SAF) and insulin sensitivity than that of VAF in non-obese Japanese diabetic subjects. Therefore, it is perhaps possible that there is different association between regional AF and insulin sensitivity between lean and obese Asian type 2 diabetes. Unfortunately, the study in a small number of obese and non-obese Chinese type 2 diabetic patients was inconclusive.[13] The objectives of the current study were to determine regional AF distribution and its relationships with insulin sensitivity and cardiovascular risk factors in lean and obese Thai type 2 diabetic patients.

MATERIALS AND METHODS

Twenty Thai type 2 diabetic women, 9 lean and 11 obese, with respective mean ages of 41.7 ± 6.3 (SD) and 48.0 ± 8.5 years, and mean body mass index (BMI) 23.5 ± 1.8 and 30.3 ± 3.7 kg/m^2, were enrolled into the study. Seven patients of the lean group and 9 of the obese group had type 2 diabetes, whereas the rest had impaired glucose tolerance by World Health Organization (WHO) criteria. One and 4 patients of the lean and the obese groups, respectively, were in menopause. Five patients of the lean group and 6 of the obese group were taking sulfonylureas and/or metformin prior to inclusion in the study. Duration of treatment of the respective groups ranged from 1 to 6 and 1 to 4 years. None was treated with insulin. Two patients had a history of hypertension. All had no recent history of major intercurrent illnesses prior to study. Patients were studied for 2 consecutive days between 8 and 11 AM after a 10-hour fast. They were advised to not undergo strenous exercise and to stop smoking and drinking alcohol for at least 24 hours prior to the study, as well as to stop their antidiabetic and antihypertensive drugs at the day of study.

On day 1, a euglycemic hyperinsulinemic clamp was performed to determine insulin sensitivity.[14] On day 2, a 75-g oral glucose tolerance test (OGTT) was performed. Blood was collected at before and 1, 2, and 3 hours after glucose ingestion via retained intravenous catheter for the measurements of glucose and insulin. Fasting blood samples were also collected for the measurements of total cholesterol (TC), triglyceride (TG), high-density lipoprotein cholesterol (HDL-C), fibrinogen, and uric acid levels. Sitting systolic (SBP) and diastolic (DBP) blood pressure were measured after an at least 15-minute rest using a mercury sphygmomanometer (Baumanometer, W.A. Baumm, Copiague, NY). TBF, total AF, and regional AF were determined.

The study was approved by the Ethic Committee of Faculty of

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Submitted January 27, 2003; acepted April 4, 2003.

Supported by grants from Department of Medicine and University research funds of Prince of Songkla University.

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0026-0495/03/5211-0043$30.00/0

doi:10.1016/S0026-0495(03)00257-9

Medicine, Prince of Songkla University. All patients gave written informed consent to participate in the study.

Euglycemic Hyperinsulinemic Clamp

Intravenous catheters were retained in antecubital vein for infusion of insulin and glucose and in contralateral dorsal hand vein for blood sampling. A primed continuous infusion of regular insulin (Actrapid HM, Novo Nordisk, Denmark) was given at a rate of 50 mU/m^2 body surface area/min from 0 to 120 minutes together with 20% dextrose solution to maintain plasma glucose at the level of 5 mmol/L throughout the clamp period. Blood samples were obtained every 5 minutes from the hand vein kept in a thermoregulated box at 55 to 60°C for determination of arterialized plasma glucose. Blood samples were also collected every 10 minutes from 60 to 120 minutes for measurement of plasma insulin levels. Insulin sensitivity was determined from the glucose infusion rate (GIR) during the last 40 minutes of the clamp and expressed as milligrams of glucose per kilogram fat free mass (FFM) per minute.

Body Composition Measurements

TBF and total AF at L1-L4 level were measured by dual energy x-ray absorptiometer (DPX-MD Lunar Corp, Madison, WI) software version 4.7. TBF was calculated by standard software program of machine, whereas AF measurement was undertaken by manually defining the area of measurement from the top of L1 to the bottom of L4. SAF and VAF areas were determined by single-slice computerized tomography scan (CT; Tomoscan AV, Philips, Japan) of the abdomen at the L4-L5 disc space level. Scanning was performed at 120 kV. Fat tissue density was determined according to flexible attenuation ranges derived from subcutaneous fat density of each individual, the method of which has been shown to be highly correlated with and may be superior to the conventional method using fixed attenuation ranges of −190 to −30 Hounsfield units.[15] SAF area was determined by subtracting the nonsubcutaneous fat from total AF area. Nonsubcutaneous AF was defined as intra-abdominal and retroperitoneal fat, including the abdominal wall muscles, paraspinal muscles, and vertebral column. VAF area was defined as intra-abdominal fat area bound by parietal peritoneum excluding the vertebral column and the paraspinal muscles. The retroperitoneum fat was included in VAF measurement.

The measurements of AF, SAF, and VAF were performed by the same operator, three times in each subject, the mean values of which were used for the study. The intra-observer coefficient of variation (CV) of each measurement was less than 3.0%.

Biochemical Analysis

Blood for plasma insulin was collected in chilled tubes, cold spun, and frozen at −80°C until analysis, all within 1 month after collection. Plasma insulin was measured by double-antibody radioimmunoassay (Diagnostic Products Corp, Los Angeles, CA) with an intra-assay CV of 0.9% to 4.7%. Plasma glucose was measured by glucose oxidase method (Synchron CX-3 Delta, Beckman Coulter, Fullerton, CA) with an inter-assay CV of 0.9% to 2.3%. TC, TG, and HDL-C were analyzed by enzymatic colorimetric method. Low-density lipoprotein cholesterol (LDL-C) was calculated from Friedewald formula. The intra- and interassay CVs of TC, TG, and HDL-C were 0.6% and 2.7%, 1.0% and 3.2%, and 1.5% and 3.2%, respectively. Fibrinogen and uric acid were respectively determined by prothrombin time (PT)-derived fibrinogen and enzymatic methods with interassay CVs 4.45% and 0.9% to 1.9%. Areas under the curve (AUCs) of glucose and insulin were calculated using the trapezoidal rule.

Statistical Analysis

Student's *t* test was used to compare means. Data that were not normally distributed were log-transformed prior to analysis. Correlation coefficients were determined by Pearson's product moment. All statistical analyses was performed using SPSS for Windows version 9 (SPSS Inc, Chicago, IL). $P < .05$ was considered statistically significant.

RESULTS

The clinical characteristics and body fat data of lean and obese type 2 diabetic women in this study are shown in Table1. Lean patients not only had lower TBF, percent TBF, and AF, but also had lower SAF and VAF than obese patients, although the ratio of VAF/SAF was not different between the groups.

In the lean group, AF ($r = -0.7663$; $P = .027$) but not TBF ($r = -0.0922$) or percent TBF ($r = 0.6007$) was correlated with GIR. With regards to regional AF, it was found that VAF was inversely correlated ($r = -0.7534$; $P = .019$), whereas SAF was positively correlated with GIR ($r = 0.7023$; $P =$

Table 1. Clinical Characteristics and Body Fat Distribution of Subjects in the Study

	Lean (n = 9)	Obese (n = 11)	*P*
Age (yr)	41.7 ± 6.3 (34-55)	48.0 ± 8.5 (30-60)	NS
Weight (kg)	58.8 ± 5.8 (51.5-69.7)	71.8 ± 10.5 (61-92.6)	.004
BMI (kg/m^2)	23.5 ± 1.8 (21.4-25.7)	30.3 ± 3.7 (26.7-36.9)	<.0001
Diabetes duration (yr)	2.0 ± 2.1 (0-6)	1.3 ± 1.7 (0-4)	NS
Fasting glucose (mmol/L)	7.4 ± 2.2 (4.4-11.9)	6.4 ± 1.4 (4.7-8.9)	NS
SBP (mm Hg)	113.7 ± 17.9 (86-145)	115.8 ± 12.9 (98-138)	NS
DBP (mm Hg)	70.8 ± 10.7 (54-85)	70.9 ± 7.1 (60-82)	NS
TBF (kg)	19.8 ± 2.0 (17.5-23.6)	29.6 ± 6.6 (22.9-39.6)	.001
%TBF	35.0 ± 2.8 (29.2-39.3)	42.7 ± 4.8 (36.5-50.2)	<.0001
AF (kg)	2.2 ± 0.4 (1.6-2.6)	3.8 ± 1.2 (2.1-5.8)	.002
SAF (cm^2)	136.6 ± 32.8 (109.0-204.0)	260.8 ± 117.6 (94.4-430.1)	.006
VAF (cm^2)	46.4 ± 24.1 (13.2-90.0)	98.2 ± 41.1 (49.0-164.4)	.004
VAF/SAF	0.37 ± 0.23 (0.07-0.81)	0.45 ± 0.34 (0.18-1.44)	NS
GIR (mg/kg · FFM/min)	5.70 ± 3.08 (2.68-12.90)	4.88 ± 2.42 (2.4-9.31)	NS

NOTE. Data are expressed as mean ± SD (range).

Abbreviations: BMI, body mass index; SBP, systolic blood pressure; DBP, diastolic blood pressure; TBF, total body fat; AF, total abdominal fat; SAF, subcutaneous abdominal fat; VAF, visceral abdominal fat; GIR, glucose infusion rate; FFM, fat free mass; NS, not significant.

.035). However, the correlation of SAF and GIR disappeared after one outlier was excluded from data analysis. VAF, but not SAF, was still strongly and inversely correlated with GIR after adjusted for percent TBF ($r = -0.8821$; $P = .009$). VAF/SAF was also inversely correlated with GIR ($r = -0.7274$; $P = .026$), but was less strong than VAF per se even after adjusted for percent TBF ($r = -0.7854$; $P = .036$).

In the obese group, TBF, percent TBF, and AF were not significantly correlated with GIR ($r = -0.2419$, 0.2891, and -0.0245, respectively). This was also true with SAF ($r = -0.030$), VAF ($r = -0.3611$), and VAF/SAF ($r = -0.3697$). However, after adjustment for percent TBF, only VAF tended to be significantly correlated with GIR ($r = -0.5820$; $P = .078$).

Since the correlations of body fat and GIR in both lean and obese patients were in the same direction, patients from both groups were combined to study the relationships of body fat distribution and cardiovascular risk. VAF was found to be significantly correlated with DBP ($r = 0.460$; $P = .041$), fasting insulin ($r = 0.636$; $P = .003$), and uric acid ($r = 0.590$; $P = .008$), whereas SAF was significantly correlated only with TG ($r = 0.506$; $P = .027$) in univariate analyses. Nevertheless, as shown in Table 2, after adjustment for percent TBF, VAF was strongly correlated with SBP, DBP, fasting insulin, and uric acid, but SAF was not correlated to any of the cardiovascular risk factors. In contrast, TBF was shown to have significant correlation with fasting and AUC of insulin, TG, and uric acid. GIR had no correlation with any of these cardiovascular risk factors after adjustment for VAF.

DISCUSSION

The results of this study demonstrated that VAF was a stronger determinant of insulin sensitivity than SAF and TBF in both lean and obese Thai type 2 diabetic women. The inverse relationship of VAF and insulin sensitivity has been consistently shown in obese subjects, both nondiabetics and diabetics, the relationship of which is stronger than that of SAF or TBF in most studies.[1,4-6,9,16] It is known that the amount of VAF plays an important role in the determination of insulin sensitivity. Since the amount of VAF positively correlates with the amount of TBF, the relationship of VAF and insulin sensitivity may not be demonstrated in lean subjects, particularly in women. The studies in lean, nondiabetic women by Tai et al from Singapore,[17] and our own study,[18] also support this idea. Most studies concerning the relationship of body fat distribution and insulin sensitivity in type 2 diabetes are performed in obese patients; studies in lean patients are relatively few. The current study confirms that VAF is the important determinant of insulin sensitivity and cardiovascular risk not only in obese but also in lean diabetic subjects. The correlation of VAF and insulin sensitivity in obese diabetic patients is consistent with several other studies. However, the strong inverse correlation of VAF and insulin sensitivity in lean patients is in contrast with the findings of Taniguchi et al in non-obese Japanese type 2 diabetic patients.[12] They reported a stronger inverse association of insulin sensitivity estimated by homeostasis model assessment with SAF than that with VAF. An inverse association of VAF and insulin sensitivity measured by a short insulin tolerance test was also demonstrated in slightly obese Chinese type 2 diabetic patients.[13] This finding is in contrast with our previous study in lean nondiabetic subjects,[18] where association of AF and insulin sensitivity could not be demonstrated. This supports the important role of VAF in determination of the insulin resistance observed in type 2 diabetes, even in lean subjects.

It is interesting to note that lean type 2 diabetic patients in this study had a degree of insulin resistance, indicated by low GIR, similar to that of obese patients despite having less TBF and AF (Table1). Insulin secretion determined by the AUC of insulin after an OGTT was also not different (data not shown). Of 9 lean patients, only 2 had GIRs of 7.96 and 12.90 mg/kg · FFM/min, while the others had GIRs that ranged from 2.68 to 5.70 mg/kg · FFM/min, comparable to the GIRs of obese patients. The higher GIRs and the lower degree of insulin resistance corresponded to the smaller VAF areas in these 2 patients. This finding is in contrast to the studies in Caucasian diabetic populations where non-obese patients were less insulin-resistant or, in other words, more insulin-sensitive than obese patients.[1,19] However, although one may not be able to extrapolate our findings to lean Thai type 2 diabetic women in general given the small number of subjects in the study, the current study does indicate that there is a subset of lean diabetic patients who have insulin resistance as severe as obese patients. This finding, if confirmed in a larger study, should have clinical relevance. Treatments that improve insulin sensitivity, not insulin secretion, should be considered as first-line therapy in this group of patients. Although this study included few patients with impaired glucose tolerance, we speculate it would not influence the results of the study since patients with impaired glucose tolerance have been demonstrated to have a degree of insulin resistance and adiposity similar to that of type 2 diabetics. It is possible that the inclusion of postmenopausal women may have had a somewhat deleterious effect on insulin sensitivity and cardiovascular risk factors, particularly in obese

Table 2. Correlation of Body Fat and Cardiovascular Risk Factors (N = 20)

Parameter	VAF*	SAF*	TBF
GIR	−0.7244§	−0.2406	−0.2270
SBP	0.5279†	−0.0659	0.0800
DBP	0.6492‡	−0.1859	0.1830
Fasting glucose	0.4165	−0.1176	−0.2250
AUC glucose	0.4018	−0.1525	−0.1980
Fasting insulin¶	0.7256§	0.0975	0.5170†
AUC insulin¶	0.3146	0.1404	0.6250‡
LDL-C	−0.0011	−0.0579	0.1790
TG¶	0.3964	0.3076	0.6680‡
HDL-C	−0.4619	−0.1715	0.0040
Fibrinogen	−0.1887	0.2634	0.3320
Uric acid	0.4963†	−0.0774	0.4900†

Abbreviations: SBP, systolic blood pressure; DBP, diastolic blood pressure; LDL-C, low-density lipoprotein cholesterol; HDL-C, high-density lipoprotein cholesterol; TG, triglyceride; VAF, visceral abdominal fat; SAF, subcutaneous abdominal fat; TBF, total body fat; GIR, glucose infusion rate; AUC, area under curves.

*Partial correlation adjusted for %TBF.

†$P < .05$, ‡$P \leq .01$, §$P \leq .001$.

¶Log-transformed prior to analysis.

patients, which is secondary, in part, to the accumulation of VAF after menopause.[19]

This study agrees and supports other studies that VAF plays a more important role than SAF or TBF in determination of cardiovascular risk factors commonly observed in type 2 diabetic patients.[11,20] What is different from other studies is that we found a strong correlation of TG with TBF, but not VAF.[5,11-13,20] We also found no correlation between HDL-C and VAF. The association of VAF with cardiovascular risk factors was consistently demonstrated in both lean and obese patients when the analysis was performed by group (data not shown). However, the association of TBF with cardiovascular risk factors could not be demonstrated in lean patients. This is possibly due to the lower level of body fat in this group of patients. The absence of a direct association between insulin sensitivity and cardiovascular risk factors independent of VAF is in contrast with the study by Haffner et al.[20] They reported significant relationships between insulin sensitivity and TG, HDL-C, fasting glucose, and plasminogen activator inhibitor-1 levels independent of waist circumference, which is the anthropometric marker of VAF. This discrepancy is possibly due in part to the small number of subjects in our study.

In conclusion, our study shows that VAF, not SAF or TBF, is a strong determinant of insulin sensitivity in both lean and obese Thai type 2 diabetic women and also determines several cardiovascular risk factors associated with type 2 diabetes.

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