Visceral Adipocyte Hypertrophy is Associated With Dyslipidemia Independent of Body Composition and Fat Distribution in Women

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OBJECTIVE—We assessed whether subcutaneous and omental adipocyte hypertrophy are related to metabolic alterations independent of body composition and fat distribution in women.

RESEARCH DESIGN AND METHODS—Mean adipocyte diameter of paired subcutaneous and omental adipose tissue samples was obtained in lean to obese women. Linear regression models predicting adipocyte size in both adipose tissue depots were computed using body composition and fat distribution measures (n = 150). In a given depot, women with larger adipocytes than predicted by the regression were considered as having adipocyte hypertrophy, whereas women with smaller adipocytes than predicted were considered as having adipocyte hyperplasia.

RESULTS—Women characterized by omental adipocyte hypertrophy had higher plasma and VLDL triglyceride levels as well as a higher total-to-HDL cholesterol ratio compared with women characterized by omental adipocyte hyperplasia ($P < 0.05$). Conversely, women characterized by subcutaneous adipocyte hypertrophy or hyperplasia showed a similar lipid profile. In logistic regression analyses, a 10% enlargement of omental adipocytes increased the risk of hypertriglyceridemia (adjusted odds ratio [OR] 4.06, $P < 0.001$) independent of body composition and fat distribution measures. A 10% increase in visceral adipocyte number also raised the risk of hypertriglyceridemia (adjusted OR 1.55, $P < 0.02$). Associations between adipocyte size and homeostasis model assessment of insulin resistance were not significant once adjusted for adiposity and body fat distribution.

CONCLUSIONS—These results suggest that omental, but not subcutaneous, adipocyte hypertrophy is associated with an altered lipid profile independent of body composition and fat distribution in women. ***Diabetes*** **60:1504–1511, 2011**

Fat tissue expansion under a positive energy imbalance relies on adipocyte hypertrophy (enlargement of existing adipocytes) and adipose tissue hyperplasia (proliferation and differentiation of preadipocytes) (1,2). Several observational studies have demonstrated that mean adipocyte sizes in abdominal subcutaneous and visceral adipose tissue are strongly associated with body composition and fat distribution measures (1,3–5). Adiposity and fat cell size are also intimately related to adipocyte function and to the metabolic alterations associated with obesity. However, factors other than adiposity and fat distribution seem to influence adipocyte size in the subcutaneous and omental adipose tissue depot (4). For example, Weyer et al. (6) have demonstrated that a significant portion of subcutaneous adipocyte size variability is explained by sequence variation in the *lamin A/C* gene, even after adjustment for body composition. Moreover, changes in adipocyte turnover rates and extracellular matrix composition may also modulate the association between adipocyte size and adiposity (6,7).

The large interindividual variability observed in adipocyte size at a given adiposity level suggests that the proneness to fat cell hypertrophy in each fat compartment may differ among individuals. Previous studies have shown that although adiposity and fat distribution are associated with several metabolic alterations, subcutaneous adipocyte size remains an independent predictor of these alterations (1,8). Specifically, enlarged subcutaneous adipocytes were associated with hyperinsulinemia and peripheral insulin resistance independent of adiposity levels (1,5,9–11). More recently, Arner et al. (1) demonstrated that subcutaneous adipocyte hypertrophy was linked to low insulin sensitivity and high insulin levels independent of body composition. This association may arise from the fact that hypertrophic adipocytes are more lipolytic, are more resistant to insulin action than small adipocytes, and have an altered adipokine secretion pattern (12–16).

Although visceral adipose tissue accumulation is known as an important predictor of metabolic alterations (17,18), previous studies did not take into account fat distribution in the association between adipocyte size and measures of glucose homeostasis or blood lipids (1,5,9–11,19). Moreover, most of these studies could not consider visceral adipose tissue cellularity (1,5,9,11). The aim of the current study was, therefore, to assess the impact of interindividual variation in abdominal subcutaneous and omental adipocyte size on measures of glucose homeostasis and blood lipid-lipoprotein levels independent of body composition and fat distribution in women. We tested the hypothesis that women characterized by adipocyte hypertrophy in either omental or subcutaneous fat, but with similar values of body composition and fat distribution, would be more likely to present metabolic alterations.

RESEARCH DESIGN AND METHODS

Subjects. The main study sample included 190 women aged 33 to 68 years recruited through the elective surgery schedule of the Gynecology Unit at the Laval University Medical Research Center, Québec, Canada. Surgeries were scheduled for total (n = 184) or subtotal (n = 6) abdominal hysterectomies sometimes accompanied by a salpingo-oophorectomy of one (n = 27) or two (n = 71) ovaries. Reasons for surgery included one or more of the following: myoma (n = 90), menorrhagia/menometrorrhagia (n = 83), endometriosis (n = 30), fibroids (n = 29), benign cyst (n = 28), incapacitating dysmenorrhea (n = 18), pelvic pain (n = 11), endometrial hyperplasia (n = 8), polyp (n = 7), pelvic adhesions (n = 5), adenomyosis (n = 3), severe premenstrual syndrome (n = 2),

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and/or ovarian thecoma (n = 1). Menopausal status determination was based on follicle-stimulating hormone levels, menstrual history questionnaires, and medical files. Thirty-three women were identified as postmenopausal, whereas the remaining women were identified as pre- or peri-menopausal (n = 157). Twenty-seven women were using hormonal replacement therapy. Severely obese women (n = 17) undergoing biliopancreatic diversion were recruited at the Laval University Cardiology and Pulmonology Institute. They were aged 47 years on average, and their BMI ranged from 39.9 to 70.5 kg/m^2. Severe obesity was defined using the BMI cutoff of the World Health Organization (WHO) (40 kg/m^2). Written informed consent was obtained from all women. The project was approved by the ethics committees of Laval University Cardiology and Pulmonology Institute and Laval University Medical Research Center.

Total adiposity and body fat distribution measurements. Within a few days before or after the surgery, measurements of total adiposity and body fat distribution were performed in most women (n = 150). Classification of normal weight, overweight, and obese women was performed using BMI cutoff points of WHO. Total body fat mass and lean body mass were determined using dual-energy X-ray absorptiometry. Abdominal subcutaneous and visceral adipose tissue cross-sectional area measures were obtained at the L4-L5 vertebrae level using a Light Speed 1.1 CT scanner (General Electric Medical Systems, Milwaukee, WI) (20,21).

Lipid profile and glucose homeostasis. Overnight fasting blood samples were drawn on the morning of the surgery. Plasma VLDL were isolated by ultracentrifugation, and the HDL fraction was obtained after precipitation of LDL in the infranatant with heparin and $MnCl_2$. The HDL_2 and HDL_3 subfractions were obtained after further precipitation of HDL_2 with dextran sulfate. Cholesterol and triglyceride levels were measured in plasma and lipoprotein fractions by enzymatic methods with a Technicon RA-500 analyzer (Bayer, Etobicoke, Canada) as previously described (22). Apolipoprotein (apo) B concentrations were measured in plasma, VLDL, or LDL fractions by the rocket immunoelectrophoretic method of Laurell (20). Glucose was measured using the glucose oxidase method and insulin was quantified by radioimmunoassay (Linco Research, St. Charles, MO). The homeostasis model assessment of insulin resistance (HOMA-IR) index was calculated using the following formula: fasting insulin (in μU/mL) × fasting glucose (in mmol/L) ÷ 22.5 (23). None of the women recruited from the gynecology unit had a previous diagnosis of diabetes or took antidiabetic drugs. However, monitoring fasting plasma glycemia on the morning of surgery revealed that 30 women had impaired fasting glycemia (>6.1 mmol/L).

Adipose tissue sampling and adipocyte isolation. Within 15 min after the beginning of the surgical procedure, paired samples of subcutaneous and omental adipose tissue were respectively collected at the site of surgical incision and the greater omentum. Samples were immediately carried to the laboratory. A portion of the tissue sample was digested 45 min at 37°C in Krebs-Ringer-Henseleit buffer supplemented with 350 units/mL of type 1 collagenase according to a modified version of the Rodbell method (21,24). Mature adipocyte suspensions were then washed three times using Krebs-Ringer-Henseleit buffer.

Adipocyte diameter and number. Mature adipocyte suspension pictures were acquired using a contrast phase microscope. Mean adipocyte diameter for each adipose tissue sample was calculated from 250 individual measurements using Scion Image software (Frederick, MD). Mean adipocyte diameter was calculated for each distribution. SD and skewness of adipocyte size distributions were not independently associated to metabolic alterations and were not included in subsequent analyses. The number of subcutaneous and visceral adipocytes at the L4-L5 vertebrae level was estimated by dividing abdominal subcutaneous and visceral adipose tissue area, respectively, by subcutaneous and omental mean cross-sectional adipocyte surface.

Statistical analyses. Nonlinear curve fit of the association between subcutaneous or omental adipocyte size and BMI was performed using a one-phase exponential equation in Prism GraphPad Software. An independent linear regression analysis was performed to predict adipocyte size in each compartment separately using measures of body composition (BMI, fat mass, and lean body mass) and fat distribution (total, subcutaneous, and visceral adipose tissue area at L4-L5 vertebrae) as well as age, menopausal status, and hormone replacement therapy. In models with nonnormally distributed residuals, the dependent variable was log-transformed. Colinearity among independent variables was assessed using inflation and condition index statistics. Models were first selected based on the Mallows' Cp statistic and further improved using the adjusted R^2 and the predicted residual sum of squares (PRESS). For each model, the study sample was stratified in two subgroups according to whether women had a positive (hypertrophy) or negative (hyperplasia) residual. In each fat depot model, adiposity and metabolic measures between subgroups were compared using t tests. Variables that were not normally distributed based on a significant Shapiro-Wilk test ($P < 0.05$) were $\log_{10}$- or Box-Cox-transformed.

Two subgroups of 54 women matched for visceral adipose tissue area and subcutaneous adipocyte size but with either small or large omental adipocytes were selected. Age and total body fat mass were also carefully matched between both subgroups. Adiposity and metabolic measures between women characterized by small or large omental adipocytes were compared using paired t tests. Multivariate logistic regression analyses were performed to predict the probability of being characterized by hypertriglyceridemia (>1.69 mmol/L) using measures of adipocyte size and number in each adipose tissue depot. The model was adjusted for body composition and fat distribution variables as well as age and menopausal status. Odds ratios (ORs) for each independent variable were computed for a 10% increase. Statistical analyses were performed using SAS (SAS Institute).

RESULTS

Subcutaneous and omental adipocyte size was measured in a sample of 207 lean to severely obese women. The curve-linear relationship between BMI and adipocyte size in each depot is shown in Fig. 1. In both fat compartments, adipocyte size increased along with BMI and tended to reach a plateau at ~130 μm in severely obese women. Taken as a whole, omental adipocytes were smaller than subcutaneous adipocytes but tended to reach similar sizes in severely obese women (BMI >40 kg/m^2). Subsequent analyses were performed in lean to moderately obese women undergoing gynecological surgery, since the relation between adipocyte size and adiposity became nonlinear in severely obese women.

Associations between regional adipocyte size and lipid profile measures were computed in a subsample of women for which data on body composition, fat distribution, and lipid profile were available (n = 150). Characteristics of these women are shown in Table 1. According to mean BMI and body composition measures, women were overweight but covered a wide range of adiposity values. We performed linear regression models to predict subcutaneous and omental adipocyte sizes using all body composition and fat distribution variables available (Table 2). Based on this analysis, lean body mass as well as subcutaneous and visceral adipose tissue area explained 30.8% of the variance in subcutaneous adipocyte size (model 1). Lean body mass, BMI, and visceral adipose tissue area explained 54.3% of the variance in omental adipocyte size (model 2).

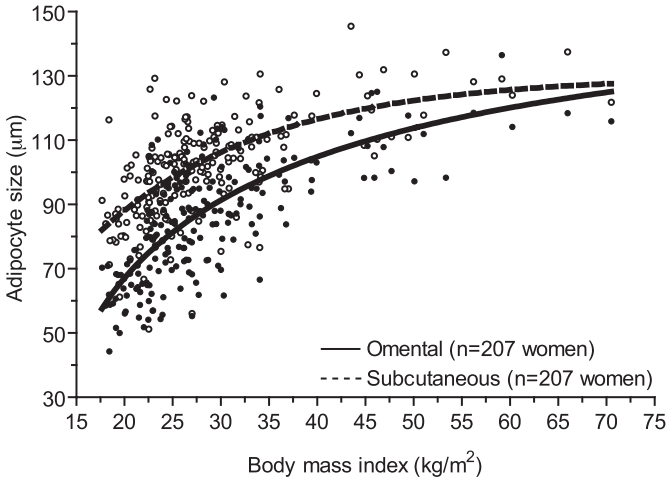

FIG. 1. Subcutaneous and omental adipocyte diameter according to BMI in women undergoing abdominal gynecologic surgery (n = 190) and biliopancreatic diversion (n = 17). Women were aged 47.7 ± 5.5 years (range 30–68.3 years) with a mean BMI of 28.5 ± 8.8 kg/m^2 (range 17.6–70.5 kg/m^2). Mean subcutaneous and omental adipocyte sizes were 101.5 ± 15.4 μm and 85.2 ± 18.6 μm, respectively.

TABLE 1
Physical and metabolic characteristics of the subsample of 150 women

Variables	Mean ± SD	Range (minimum–maximum)
Anthropometrics		
Age (years)	47.1 ± 5.1	36.6–68.3
BMI (kg/m^2)	26.7 ± 4.8	18.4–39.4
Body fat mass (kg)	26.5 ± 9.3	5.8–53.5
Lean body mass (kg)	40.8 ± 6.6	25.3–56.4
Adipose tissue area (cm^2)		
Total	413 ± 163	87–992
Subcutaneous	321 ± 131	51–759
Visceral	93 ± 46	19–266
Lipid profile (mmol/L)		
Plasma triglycerides	1.32 ± 0.58	0.47–3.12
VLDL triglycerides	0.80 ± 0.49	0.12–2.18
Total cholesterol	5.04 ± 0.95	2.62–7.52
VLDL cholesterol	0.47 ± 0.30	0.09–1.92
LDL cholesterol	3.13 ± 0.86	1.07–5.69
HDL cholesterol	1.44 ± 0.36	0.63–2.68
Apolipoprotein B (g/L)	0.95 ± 0.25	0.39–1.61
Total-to-HDL cholesterol ratio	3.70 ± 1.13	1.66–9.29
Glucose homeostasis		
Fasting glucose (mmol/L)†	5.67 ± 0.63	4.80–8.04
Fasting insulin (pmol/L)††	9.5 ± 5.5	3.3–27.6
HOMA-IR index†††	2.52 ± 1.64	0.72–8.85
Adipocyte diameter (μm)		
Subcutaneous	99.7 ± 12.8	61.2–130.7
Omental	83.3 ± 16.6	44.3–120.5

†n = 138; ††n = 141; †††n = 135.

TABLE 2
Linear regression analyses predicting omental and subcutaneous adipocyte size in women

Variables*	Partial ($R^2 \times 100$)	Total ($R^2 \times 100$)	P value
Model 1			
Dependent			
Subcutaneous adipocyte size			
Independent			
Visceral adipose tissue area	24.6	30.8	<0.0001
Subcutaneous AT area	4.8		<0.005
Lean body mass	1.4		0.09
Model 2			
Dependent			
Omental adipocyte size†			
Independent			
Visceral adipose tissue area	47.4	54.3	<0.0001
BMI	3.5		<0.005
Lean body mass	3.4		<0.005

*Regression models included BMI, body fat mass, lean body mass, subcutaneous adipose tissue area, visceral adipose tissue area, age, menopausal status, and hormone replacement therapy. Models were selected based on the Mallows' Cp, adjusted R^2, and PRESS statistics; colinearity among independent variables was assessed through inflation and condition index statistics. †Log-transformed variables; n = 150. AT: adipose tissue.

According to these models, a large percentage of the variance in adipocyte size remained unexplained despite considering body composition and fat distribution measures. To assess the impact of adipocyte size variation on the metabolic profile independent of body composition and fat distribution, we stratified the study sample according to the difference between measured and predicted adipocyte size in each model. Women with larger adipocytes than predicted by the regression model (positive residual) were identified as hypertrophic, whereas women with smaller adipocytes than predicted by the model were identified as hyperplasic (negative residual) in the corresponding adipose tissue compartment. By design, stratifications performed using each model generated subgroups of women characterized by identical body composition and fat distribution values.

Figure 2 shows adipocyte size and number in each adipose tissue compartment according to the presence of hypertrophy or hyperplasia in subcutaneous (*left*) or omental (*right*) adipose tissue. Women characterized by hypertrophic subcutaneous adipocytes had fewer but larger subcutaneous adipocytes than women with hyperplasic subcutaneous adipocytes ($P < 0.0001$). Women characterized by subcutaneous adipocyte hypertrophy also had slightly larger omental adipocytes, but their cell number remained similar to that of women characterized by subcutaneous adipocyte hyperplasia ($P < 0.05$). Similarly, larger but fewer omental adipocytes were observed in women characterized by omental adipocyte hypertrophy compared with women characterized by omental adipocyte hyperplasia ($P < 0.0001$). Although the number of subcutaneous adipocytes was similar in both groups derived from model 2, slightly larger subcutaneous adipocytes were observed in women characterized by omental adipocyte hypertrophy ($P < 0.05$).

No significant difference in blood lipids was observed between women characterized by subcutaneous adipocyte hypertrophy versus subcutaneous adipocyte hyperplasia (Fig. 3, *left*). On the other hand, women characterized by omental adipocyte hypertrophy had higher levels of plasma triglycerides, higher VLDL triglycerides, and higher VLDL cholesterol levels compared with women characterized by omental adipocyte hyperplasia (Fig. 3*A–C*, *right*). The ratio of total cholesterol to HDL cholesterol was significantly higher in women characterized by omental adipocyte hypertrophy compared with women characterized by omental adipocyte hyperplasia (Fig. 3*D*, *right*). Increased triglyceride and cholesterol content in the VLDL fraction may suggest increased particle size (10.89 ± 3.85 vs. 9.27 ± 3.97 mmol of lipids/g of ApoB, $P < 0.01$) rather than increased particle number as estimated by VLDL-ApoB (0.13 ± 0.06 vs. 0.12 ± 0.07 g/L, NS).

Subdivision of women in three subgroups according to the SD of the linear regression model residuals generated results that are comparable with the two-group analyses. Specifically, women with adipocyte sizes inside of 1 SD around the mean showed an intermediate phenotype for blood lipid levels compared with women above or below 1 SD of the regression residuals (data not shown).

No significant difference was observed in fasting glucose (5.75 ± 0.65 vs. 5.60 ± 0.59, NS) and insulin levels (10.15 ± 5.93 vs. 8.88 ± 5.05, NS) as well as in HOMA-IR index (2.68 ± 1.71 vs. 2.34 ± 1.60, NS) among women characterized by omental adipocyte hypertrophy or hyperplasia. Similar results were observed between women characterized by subcutaneous adipocyte hypertrophy or hyperplasia. However, when the linear regression analyses were performed using a single measure of total adiposity (i.e., fat

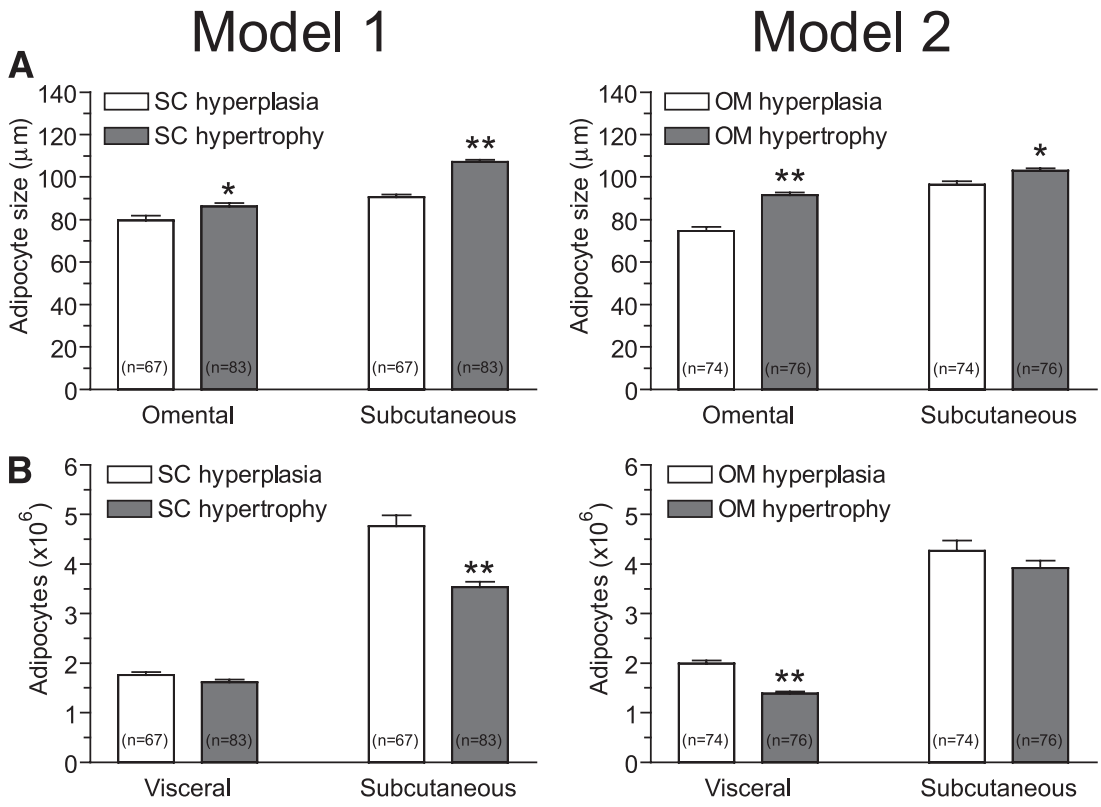

FIG. 2. Adipose tissue cellularity in women characterized by hypertrophic or hyperplasic subcutaneous and omental adipocytes. Adipocyte diameter (*A*) and adipocyte number at the L4-L5 vertebrae level (*B*) are shown for subcutaneous (SC; model 1; *n* = 150) and omental (OM; model 2; *n* = 150) adipose tissue cellularity stratifications. Values are mean ± SEM. **P* < 0.05; ***P* < 0.01.

mass) to predict adipocyte size, we observed a trend for higher fasting glucose (5.75 ± 0.64 vs. 5.57 ± 0.59, $P = 0.09$), significantly higher fasting insulin levels (10.9 ± 6.3 vs. 7.8 ± 3.8, $P < 0.005$), as well as a higher HOMA-IR index (2. 09 ± 1.83 vs. 2.00 ± 1.16, $P < 0.001$) in women characterized by subcutaneous adipocyte hypertrophy compared with women characterized by subcutaneous adipocyte hyperplasia.

To confirm results of the regression analyses, subgroups of women matched for fat mass, visceral adipose tissue area, and subcutaneous adipocyte size but with either small or large omental adipocytes were compared (Table 3). By design, both subgroups had a similar body composition and fat distribution. Although subcutaneous adipocyte size and number were comparable, women with large omental adipocytes had a lower number of visceral adipocytes. Women with large omental adipocytes had higher levels of plasma triglycerides, of VLDL triglycerides, and of VLDL cholesterol. A trend for lower HDL cholesterol was observed in women with large omental adipocytes. The lower HDL cholesterol value was explained by significantly lower cholesterol content in HDL_2, but not in HDL_3 lipoproteins, in women with larger omental adipocytes. Moreover, the total cholesterol-to-HDL-cholesterol ratio tended to be higher in women with large omental adipocytes compared with women with small omental adipocytes. Fasting glucose and insulin concentrations as well as the HOMA-IR index were similar between women with large versus small omental adipocytes (Table 3).

The risk of hypertriglyceridemia, defined by triglyceride levels greater than 1.69 mmol/L in women, was assessed using logistic regression analyses (Table 4). When adipocyte size and number in each fat depot were included in the model, the risk of hypertriglyceridemia was associated with adipose tissue cellularity measures of the visceral, but not the subcutaneous, fat compartment. Indeed, the risk of hypertriglyceridemia was significantly higher in women with 10% larger omental adipocytes (OR 2.08 [95% CI 1.48–3.04]) and with 10% more adipocytes in visceral adipose tissue (OR 1.16 [1.03–1.30]). After adjustment for BMI, body fat mass, lean body mass, subcutaneous adipose tissue area, visceral adipose tissue area, age, and menopausal status, omental adipocyte size (adjusted OR 4.06 [1.92–9.97]) and number (adjusted OR 1.55 [1.12–2.29]) remained independent predictors of hypertriglyceridemia in women.

DISCUSSION

We designed this study to assess whether subcutaneous and omental adipose tissue cellularity measures were related to metabolic alterations independent of body composition and fat distribution in women. Using linear regression analyses, women were subdivided in two groups having lower-than-predicted (hyperplasia) or higher-than-predicted (hypertrophy) adipocyte sizes in subcutaneous and omental adipose tissue. Women characterized by omental adipocyte hypertrophy presented a deleterious lipid profile compared with women characterized by omental adipocyte hyperplasia. These alterations in the lipid profile were, by design, independent of differences in body composition and fat distribution. In contrast, lipid profiles were similar in women characterized by subcutaneous adipocyte hypertrophy versus subcutaneous adipocyte hyperplasia. In logistic regression analyses including body composition and

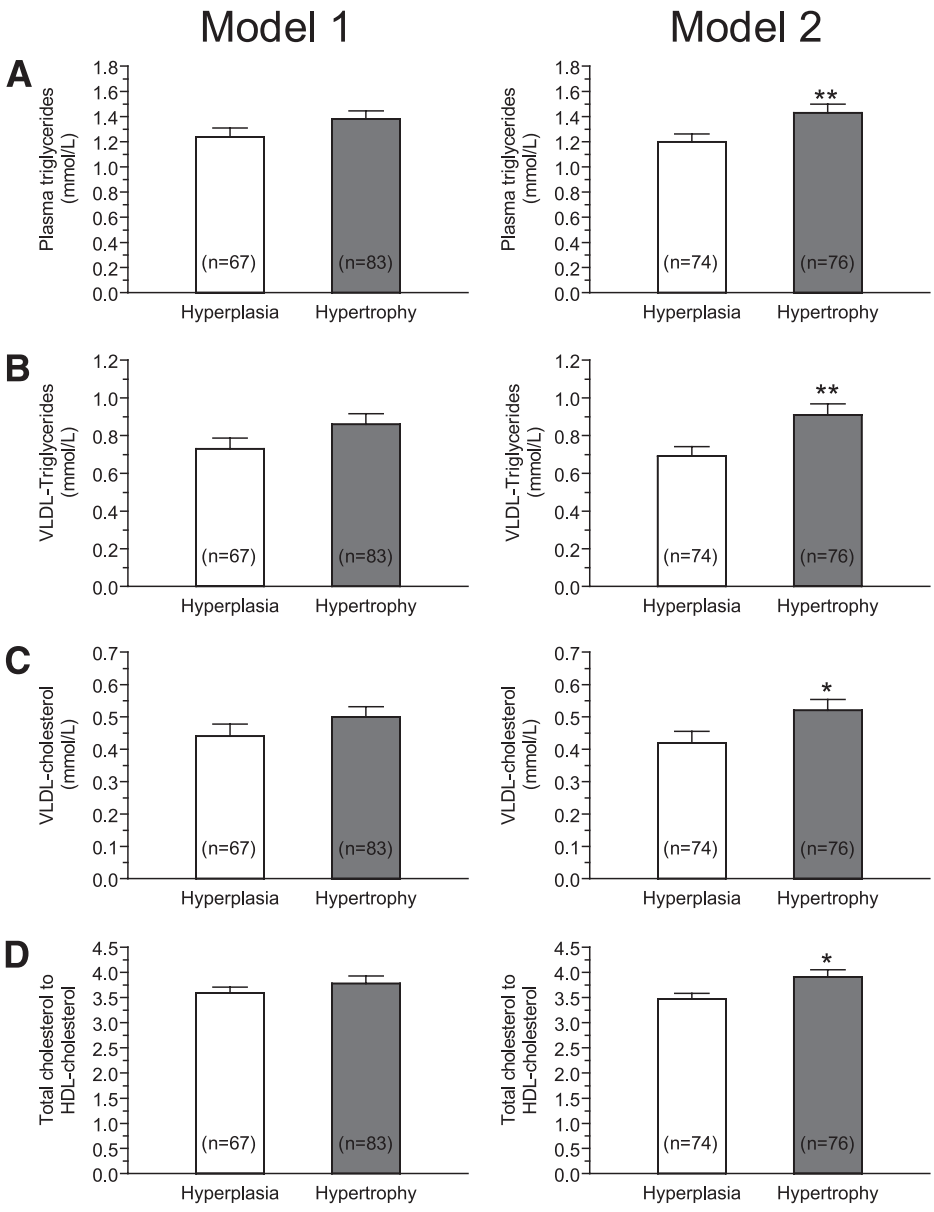

FIG. 3. Lipid profile in women characterized by hypertrophic or hyperplasic subcutaneous (model 1; n = 150) and omental (model 2; n = 150) adipocytes. Overnight fast values of plasma triglyceride (*A*) (mmol/L), VLDL triglyceride (*B*) (mmol/L), VLDL cholesterol (*C*) (mmol/L), and total-to-HDL cholesterol (*D*) are shown. Values are mean ± SEM. *$P < 0.05$; **$P < 0.01$.

fat distribution measures, we estimated that a 10% enlargement of omental adipocytes increased the risk of hypertriglyceridemia by more than fourfold, whereas enlarged subcutaneous adipocyte size failed to significantly alter the risk of hypertriglyceridemia in women. To a lower extent, a 10% increase in the number of visceral, but not subcutaneous, adipocytes multiplied the risk of hypertriglyceridemia in women by 1.55-fold. These results suggest that omental fat cell size and number predict lipid profile alterations independent of body composition and fat distribution in women. Despite the fact that total and visceral adiposity are known modulators of the lipid profile, we show here for the first time that the extent of omental adipocyte hypertrophy may influence this relation.

Previous studies reported that subcutaneous adipocyte hypertrophy was associated with alterations in glucose homeostasis (1,5,9–11). Elegant studies from Arner et al. (1) on adipocyte hypertrophy and adipocyte turnover showed that individuals characterized by larger subcutaneous adipocytes had higher fasting insulin levels and HOMA-IR index. In the current study, all measures of body composition and fat distribution as well as age and menopausal status were used to prepare fully adjusted predictive models for subcutaneous and omental adipocyte size. Using these models, we failed to observe alterations in glucose homeostasis measures in women characterized by adipocyte hypertrophy in subcutaneous or omental adipose tissue. On the other hand, linear regression analyses predicting adipocyte size performed using only one measure of total adiposity (e.g., fat mass) revealed glucose homeostasis alterations in women characterized by subcutaneous adipocyte hypertrophy compared with women characterized by subcutaneous adipocyte hyperplasia. These results suggest that associations of glucose homeostasis

TABLE 3
Characteristics of women matched for visceral adipose tissue area and subcutaneous adipocyte size but with either small or large omental adipocytes (n = 54)

Variables	Omental adipocytes		P value
	Small	Large	
Anthropometrics			
Age (years)	47.1 ± 5.7	47.4 ± 5.2	NS
BMI (kg/m^2)	28.2 ± 6.3	27.2 ± 4.8	NS
Fat mass (kg)	28.8 ± 10.7	27.6 ± 9.1	NS
Body lean mass (kg)	42.0 ± 6.7	40.9 ± 6.9	NS
Adipose tissue area (cm^2)			
Total	446 ± 179	431 ± 159	NS
Subcutaneous	350 ± 147	334 ± 123	NS
Visceral	101 ± 50	101 ± 51	NS
Adipocyte diameter (μm)			
Subcutaneous	100.9 ± 12.2	100.4 ± 10.4	NS
Omental	77.1 ± 13.5	93.6 ± 13.7	<0.0001
Number of adipocytes L4-L5 (×10^6)			
Subcutaneous depots	4.39 ± 1.71	4.21 ± 1.52	NS
Visceral depots	2.06 ± 0.53	1.40 ± 0.42	<0.0001
Lipid profile (mmol/L)			
Plasma triglycerides	1.25 ± 0.48	1.47 ± 0.66	<0.01
VLDL triglycerides	0.74 ± 0.40	0.93 ± 0.56	<0.01
Total cholesterol	5.05 ± 0.96	5.13 ± 0.98	NS
VLDL cholesterol	0.44 ± 0.23	0.55 ± 0.37	<0.05
LDL cholesterol	3.13 ± 0.94	3.20 ± 0.87	NS
HDL cholesterol	1.48 ± 0.36	1.38 ± 0.31	0.08
HDL_2 cholesterol	0.61 ± 0.27	0.53 ± 0.20	<0.05
HDL_3 cholesterol	0.87 ± 0.18	0.85 ± 0.18	NS
Apolipoprotein B (g/L)	0.94 ± 0.25	0.99 ± 0.26	NS
Total-to-HDL cholesterol ratio	3.60 ± 1.04	3.92 ± 1.30	0.07
Glucose homeostatsis			
Fasting glucose (mmol/L)†	5.62 ± 0.63	5.86 ± 0.64	NS
Fasting insulin (pmol/L)††	9.8 ± 7.7	10.5 ± 5.8	NS
HOMA-IR index†††	2.62 ± 2.22	2.85 ± 1.71	NS

†n = 46; ††n = 48; †††n = 43.

with subcutaneous adipocyte hypertrophy, as observed by others (1,5,9–11), may arise from differences in abdominal fat distribution rather than differences in subcutaneous adipose tissue cellularity. Lack of statistical adjustment for fat distribution measures in previous studies may explain discrepancies regarding glucose homeostasis alterations in women characterized by hypertrophic subcutaneous adipocytes (1,5,9–11).

To our knowledge, we are the first to report an independent association between omental adipocyte hypertrophy and alterations of the lipid profile in women. With the exception of two studies (10,19), most authors assessed the association between adipocyte size and metabolic parameters using only measures from subcutaneous adipose tissue (1,3,5,11). These studies reported that enlarged subcutaneous adipocytes were associated with insulin resistance independent of obesity itself (1,5,11). In contrast, Mundi et al. (3) observed that insulin and triglyceride levels were slightly better predicted by body composition measures than subcutaneous adipocyte size. Therefore, they dismissed the individual effect of subcutaneous adipocyte size on metabolic parameters in men and women (3). Subcutaneous and omental adipocyte size measures are closely related (4,25), and part of the effect of omental adipocyte hypertrophy may be indirectly related to subcutaneous adipocyte size. However, lack of visceral adipocyte size measures in most of these studies may explain discrepancies observed between our analysis and previous literature (1,3,5).

We corroborate reports showing that body composition and fat distribution measures largely predict subcutaneous and omental adipocyte size (4). However, after adjustment for all anthropometric measures available, interindividual variability observed in subcutaneous and omental adipose tissue was not completely explained. This observation is consistent with the hypothesis that adipocyte size is regulated by factors that are independent of variations in body composition and fat distribution (4). Bergman et al. (26) suggested that fat accumulation occurs primarily in the visceral adipose tissue depot and fat storage then spills over in the subcutaneous adipose tissue depot, whereas others have proposed that subcutaneous fat is a primary compartment (27,28). In any case, lipid uptake and lipolysis rates in each fat depot appear as dynamic processes. Drolet et al. (2) observed that hyperplasia is predominant in the subcutaneous adipose tissue, whereas adipocyte hypertrophy is present in both adipose tissue depots. Because less than 10% of mature adipocytes are renewed each year in subcutaneous adipose tissue (29), cell hyperplasia may represent a long-term adipose tissue adaptation to face excess energy intake. Conversely, the predominance of cell hypertrophy in visceral adipose tissue may suggest that it is a short-term storage compartment. Increased adipose tissue storage capacity through peroxisome proliferator–activated receptor-γ agonist–induced hyperplasia has been shown to promote fat redistribution and to reduce visceral fat accumulation (30). These observations show that increasing adipogenic capacity in subcutaneous adipose tissue may

TABLE 4
Multivariate logistic regression analysis for triglyceride greater than 1.69 mmol/L

Independent variables (unit 10%)	Unadjusted		Adjusted*		P value
	OR	95% CI	OR	95% CI	
Omental adipose tissue depot					
Adipocyte size (8.3 μm)	2.08	1.48–3.04	4.06	1.92–9.97	<0.001
Number of adipocytes (1.7 × 10^5)	1.16	1.03–1.30	1.55	1.12–2.29	<0.02
Subcutaneous adipose tissue depot					
Adipocyte size (9.9 μm)	1.46	0.87–2.53	0.76	0.18–2.81	NS
Number of adipocytes (4.2 × 10^5)	1.05	0.91–1.22	0.72	0.37–1.26	NS

*Logistic regression model was adjusted for body fat mass (NS), subcutaneous adipose tissue area (NS), visceral adipose tissue area (P = 0.07), free fat mass (NS), BMI (NS), age (NS), menopausal status (NS), and hormone replacement therapy (NS); n = 150.

attenuate visceral adipocyte hypertrophy and related alterations.

Although this study was not designed to investigate the mechanisms involved in the association of adipocyte hypertrophy and metabolic alterations, several hypotheses may be put forward. Involvement of visceral, but not subcutaneous, adipose tissue cellularity could indirectly reinforce the role of excess free fatty acids (31). Indeed, previous studies have reported that large adipocytes showed alterations in lipolysis, insulin sensitivity, and adipokine secretion compared with smaller adipocytes from the same individual (12,13,15,16). Although visceral adipose tissue is not believed to be a major source for the circulating free fatty acid pool, the contribution of visceral adipose tissue lipolysis to hepatic free fatty acid delivery is positively associated to visceral fat accumulation (32). Increased responsiveness to β-adrenergic agonist stimulation (33–35) and decreased sensitivity to insulin suppression of lipolysis (36,37) in hypertrophic omental adipocytes could possibly impact splanchnic free fatty acid levels, especially in the visceral obese. Increased free fatty acid delivery to the liver was suggested to increased triglyceride-rich VLDL production, which is associated with small, dense LDL particles as well as with lower HDL cholesterol levels (38,39). The altered lipid profile observed in women characterized by omental adipocyte hypertrophy is, therefore, consistent with this hypothesis. An altered adipokine secretion pattern, as observed with large adipocytes, may also influence local adipose tissue lipid metabolism and generate abnormal adipose-derived signaling through the portal vein (15). Moreover, reduced cellular stability of large adipocytes may increase the risk of cell rupture in hypertrophic adipose tissue (40). Chronic, low-grade inflammation provoked by adipocyte death may then contribute to the metabolic alterations associated with obesity (41).

Results of this study are strengthened by the use of extensive measures of adiposity and body fat distribution together with characterization of subcutaneous and omental adipose tissue cellularity. Moreover, the absence of differences in body composition and fat distribution measures between women characterized by hypertrophy and hyperplasia supports the validity and the strength of the methodology used. We suggest that this stratification is adequate to assess metabolic alterations associated to subcutaneous or omental hypertrophy, independent of body composition and fat distribution. However, some limitations in the study design should be acknowledged. The present analyses are based on cross-sectional data, and it is hazardous to conclude cause-and-effect relationships between visceral adipocyte hypertrophy and the development of metabolic alterations in women. The difficulty to recruit lean to moderately obese men undergoing abdominal surgery limits our capacity to perform a comparable study in men. Most previous studies (1,3,5,9) included both men and women, but only subcutaneous adipose tissue cellularity was assessed. This study was not designed to assess the involvement of very small adipocytes in metabolic alterations since we used collagenase-digested adipose tissue samples. We could neither support nor challenge the previous report by McLaughlin et al. (42) suggesting that the proportion of very small adipocytes is associated with metabolic alterations. Finally, because we only had access to omental and not mesenteric adipose tissue, visceral adipocyte numbers were computed using omental adipocyte surface. We assumed that mean adipocyte sizes in each visceral adipose tissue depot are strongly correlated, as previously observed for the subcutaneous and visceral adipose tissue depots (4).

During the revision process of this article, Hoffstedt et al. (43) published a similar study reporting that visceral adipose tissue hypertrophy is associated with dyslipidemia. Our study is generally concordant with these results. However, they show that subcutaneous adipocyte hypertrophy was independently associated with glucose homeostasis. The recruitment of morbidly obese women and our use of detailed assessment of body fat distribution may account for these discrepancies.

In conclusion, our results support the hypothesis that omental adipose tissue cellularity is an important predictor of hypertriglyceridemia in women. We demonstrate that the association between omental adipocyte hypertrophy and lipid profile alterations is independent of differences in subcutaneous adipose tissue cellularity, body composition, and fat distribution in women.

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A.V. designed the study, performed the analyses, and wrote the manuscript. M.C.-J. performed measurements of adipocyte sizes and critically reviewed the manuscript. S.N. and P.Y.L. contributed to the clinical aspects of the study and/or critically reviewed the manuscript. A.T. designed the study and revised the manuscript.

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