

Does Measurement Site for Visceral and Abdominal Subcutaneous Adipose Tissue Alter Associations With the Metabolic Syndrome?

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OBJECTIVE — To determine whether the associations between visceral adipose tissue (VAT), abdominal subcutaneous adipose tissue (ASAT), and the metabolic syndrome are altered depending on measurement site for VAT and ASAT and the definition used to identify the metabolic syndrome.

RESEARCH DESIGN AND METHODS — Total VAT and ASAT volume was derived using ~37 contiguous computed tomography (CT) images from T10–T11 to L5–S1 in 85 men. CT images obtained at eight intervertebral locations (e.g., L4–L5, L3–L4, etc.) were used to determine the associations between partial volumes (single images) and metabolic syndrome. Metabolic syndrome was defined using the National Cholesterol Education Program (NCEP) and International Diabetes Federation (IDF) criteria. Logistic regression was used to calculate the odds ratio (OR) per SD increase in adipose tissue.

RESULTS — For total and all partial volumes, VAT was more strongly associated with metabolic syndrome than ASAT independent of metabolic syndrome criteria. The OR (per SD) for NCEP metabolic syndrome was higher for total VAT volume (OR = 7.26) and for the partial volumes at T12–L1 (7.46) and L1–L2 (8.77) than those at the L4–L5 level (3.94). The OR for metabolic syndrome (~2.6) was not substantially different among the ASAT measures. A similar pattern of association was observed using the IDF metabolic syndrome criteria.

CONCLUSIONS — The measurement site for VAT, but not for ASAT, has a substantial influence on the magnitude of the association with both metabolic syndrome definitions. However, because VAT remained significantly associated with metabolic syndrome regardless of measurement site, the clinical interpretation was unaltered by measurement protocol or metabolic syndrome definition.

Diabetes Care 29:679–684, 2006

Mounting evidence suggests that the metabolic syndrome, generally characterized using the National Cholesterol Education Program (NCEP) criteria, is a powerful predictor of morbidity and mortality (1). Because abdominal obesity is an important component of the NCEP metabolic syndrome definition (2), researchers have investi-

gated the relationship between visceral adipose tissue (VAT) and abdominal subcutaneous adipose tissue (ASAT) with metabolic syndrome (3,4). Carr et al. (3) and Goodpaster et al. (4) report that VAT is associated with an increased incidence of metabolic syndrome. Conversely, ASAT is not consistently reported to be a significant correlate of metabolic syn-

drome (4) or its individual components (3). This may be due in part to variation in the measurement site selected for quantifying abdominal adipose tissue. Although L4–L5 is the most commonly used landmark for measuring VAT and ASAT, there is reason to believe that L4–L5 is not the ideal site for quantifying abdominal adipose tissue or for predicting obesity-related metabolic risk. Recent studies report that a single image in the upper abdomen (i.e., at L1–L2 or L2–L3) is a more suitable surrogate for total VAT (5–7) and ASAT volume (7) than an image at L4–L5. If one assumes that the ability to predict the total VAT or ASAT volume will translate to a corresponding ability to predict metabolic syndrome, then VAT and ASAT volumes obtained at L1–L2 or L2–L3 may be better predictors of metabolic syndrome than measurements obtained at L4–L5. Further, there is evidence to suggest that a greater deposition of the more metabolically active visceral adipocytes within the omental and mesenteric depots is located within the region of L1–L2 to L3–L4 (8,9). Taken together these observations suggest that VAT measures in the upper abdomen should have stronger associations with metabolic syndrome than measures in the lower abdomen. No study has systematically determined the measurement site for VAT and ASAT that is optimal for determination of metabolic syndrome. Further, although several criteria exist for identifying metabolic syndrome (10), the NCEP (11) and International Diabetes Federation (IDF) (12) criteria are the most readily used in clinical settings. Unknown is whether the criteria used to identify metabolic syndrome alters the association with VAT and ASAT. Hence, the primary purpose of this study was to determine whether the associations between VAT, ASAT, and metabolic syndrome were altered depending on the site of measurement for VAT and ASAT and/or the criteria used to identify metabolic syndrome.

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Received for publication 11 August 2005 and accepted in revised form 22 November 2005.

Abbreviations: ASAT, abdominal subcutaneous adipose tissue; CT, computed tomography; IDF, International Diabetes Federation; NCEP, National Cholesterol Education Program; SEE, standard error of estimates; VAT, visceral adipose tissue.

A table elsewhere in this issue shows conventional and Système International (SI) units and conversion factors for many substances.

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Table 1—Subject characteristics for 85 men with and without metabolic syndrome according to the NCEP and IDF definitions

	NCEP definition		IDF definition	
	No MetS	MetS	No MetS	MetS
<i>n</i>	71	14	61	24
Age (years)	50.3 ± 8.1	54.4 ± 6.1*	49.4 ± 8.1	55.0 ± 6.1*
BMI (kg/m ²)	26.3 ± 3.2	30.7 ± 3.0*	25.8 ± 3.0	30.1 ± 3.0*
Waist circumference (cm)	93.0 ± 9.7	106.3 ± 6.1*	91.5 ± 9.3	104.6 ± 6.4*
VAT volume (l)	2.7 ± 1.3	4.8 ± 1.2*	2.5 ± 1.2	4.4 ± 1.3*
ASAT volume (l)	3.5 ± 1.3	4.7 ± 0.9*	3.3 ± 1.2	4.6 ± 1.2*
VAT area L4–L5 (cm ²)	126.8 ± 54.1	200.8 ± 52.4*	122.3 ± 54.5	181.5 ± 53.7*
ASAT area L4–L5 (cm ²)	211.1 ± 75.5	284.9 ± 56.5*	202.0 ± 72.2	277.5 ± 63.4*
Fasting glucose (mg/dl)	100.3 ± 8.9	107.6 ± 8.7*	99.6 ± 9.3	106.3 ± 7.3
Fasting triglycerides (mg/dl)	135.2 ± 75.1	164.6 ± 65.7	136.2 ± 78.6	131.0 ± 54.6
HDL cholesterol (mg/dl)	47.9 ± 10.1	37.7 ± 6.9*	47.3 ± 10.4	43.5 ± 9.8*
Systolic blood pressure (mmHg)†	122.5 ± 12.6	138.4 ± 14.7*	121.6 ± 12.5	134.0 ± 14.5*

Data are means ± SD. *Significantly different from no metabolic syndrome (MetS). †*n* = 80.

RESEARCH DESIGN AND METHODS

Subjects consisted of a subset of 85 Caucasian men selected from a cohort of men who had received an abdominal computed tomography (CT) scan as part of a preventive medicine diagnostic examination at the Cooper Clinic in Dallas, Texas, as described elsewhere (13). All subjects gave their informed written consent before participation in the examination, and the study was reviewed and approved annually by The Cooper Institute Institutional Review Board.

Biochemistry analyses

Fasting total serum triglycerides, HDL cholesterol, and glucose were determined using automated methods in a laboratory that participates in and meets quality control standards of the Centers for Disease Control and Prevention Lipid Standardization Program. Metabolic syndrome was defined according to the criteria established by the NCEP Adult Treatment Panel III (11) and IDF (12). Participants were classified as having NCEP metabolic syndrome if they had three or more of the following five risk factors: 1) high systolic blood pressure ≥ 130 mmHg or the use of antihypertensive medication; 2) abdominal obesity (waist circumference > 102 cm); 3) high triglyceride (≥ 150 mg/ml); 4) low HDL cholesterol (< 40 mg/ml); and 5) high fasting plasma glucose (≥ 110 mg/ml). Participants were classified as having IDF metabolic syndrome if they had central obesity (waist circumference ≥ 94 cm) and two or more of the following four risk factors: 1) high systolic blood pressure ≥ 130 mmHg or the use of antihypertensive medication; 2) high triglyceride (≥ 150 mg/ml); 3) low HDL cholesterol (< 40 mg/ml); and 4) high fasting plasma glucose (≥ 100 mg/ml).

eride (≥ 150 mg/ml); 3) low HDL cholesterol (< 40 mg/ml); and 4) high fasting plasma glucose (≥ 100 mg/ml).

Measurement of abdominal adipose tissue by CT

Axial images of the abdominal region were obtained in an electron beam CT scanner (Imatron; General Electric, Milwaukee, WI) using a standard protocol (14). Subjects were examined in a supine position with their arms extended above their heads; ~ 40 contiguous transverse images (6 mm thickness, 0 mm interspace) were acquired from the midregion of the iliac crest to the caudal region of the heart. Images were obtained using 130 kV and 630 mA with a 480 mm field of view and a 512×512 matrix, resulting in a pixel size of 0.7813 mm^2 . The CT data obtained in Dallas were transferred electronically to Queen's University for analysis using specially designed image analysis software (Tomovision, Montreal, Canada) as previously described (13). The interobserver reliability errors for VAT and ASAT measurements for two observers' analyses of the same L4–L5 image ($n = 40$) were ~ 3 and $\sim 1\%$, respectively (15).

Determination and calculation of abdominal adipose tissue partial and total volumes

For each subject a single axial image that most closely approximated the center of the intervertebral space or vertebral body was identified for L4–L5, L3–L4, etc., using known anatomical landmarks (16). The partial volumes (cubic centimeters) of adipose tissue were calculated as the adipose tissue areas (square centimeters)

at the eight intervertebral spaces within the region of T10–T11 to L5–S1 multiplied by the slice thickness (6 mm). Total VAT and ASAT volume (criterion method) was determined from the sum of all the adipose tissue areas (square centimeters) within the region of T10–T11 to L5–S1 multiplied by the slice thickness (6 mm) (~ 37 of the 40 contiguous images acquired).

Statistical analyses

Group data are presented as means ± SD. Pearson's correlations and linear regression analyses were performed to determine the relationship between the criterion measure of abdominal adipose tissue volume and the regional adipose tissue areas. The strength of the correlations was compared by using the Hotelling method (17). The standard error of estimates (SEE) was compared using Pitman's test (18). Logistic regression was used to calculate the odds ratios (OR) for metabolic syndrome. The ORs were expressed per SD increase to facilitate comparisons between adipose tissue variables. To examine these associations in overweight or obese men who are at increased risk for metabolic disorders, we analyzed data from men with a BMI $\geq 27 \text{ kg/m}^2$ ($n = 40$). All statistical procedures were performed using SAS v8.

RESULTS— The subject characteristics are listed in Table 1. The pattern of VAT and ASAT distribution within the abdomen is shown in Fig. 1. The relationships between each of the partial volumes obtained at the intervertebral spaces between T10–T11 and L5–S1 with the total VAT and ASAT volume are provided in

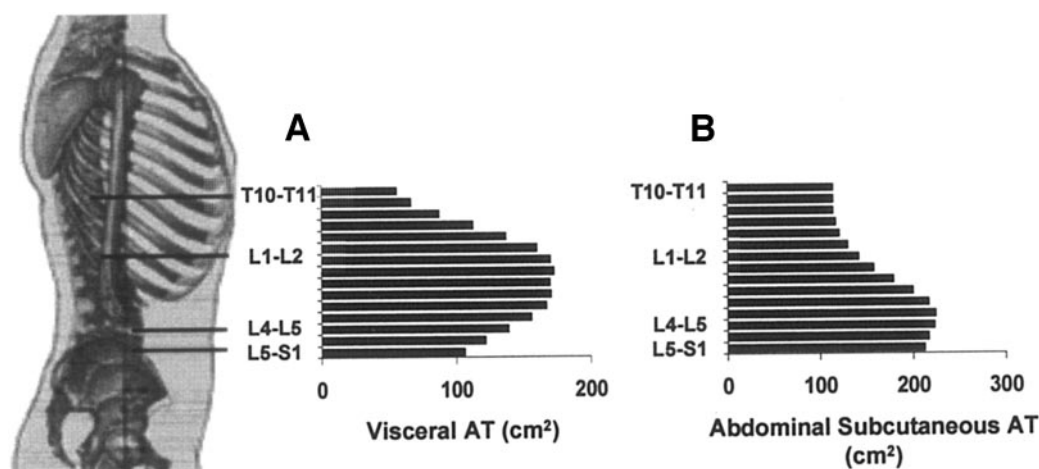


Figure 1—VAT (A) and ASAT (B) deposition at various measurement sites across the abdomen in 85 men.

Table 2. As expected, all the VAT and ASAT partial volumes derived from single images were significantly correlated with the respective total volumes for VAT ($R^2 = 0.65$ – 0.96) and ASAT ($R^2 = 0.80$ – 0.96), respectively. For VAT, measures at L1–L2 and L2–L3 had significantly higher correlation coefficients with total volume and lower SEEs than all the other images ($P < 0.05$). For subcutaneous adipose tissue, measures at L2–L3 had a significantly higher correlation coefficient with total volume and lower SEE than all the other images ($P < 0.05$) with the exception of L1–L2 ($P > 0.10$).

The metabolic syndrome was present in 16.5 and 28.2% of the sample as defined by NCEP and IDF, respectively. Waist circumference was significantly associated with both NCEP (OR = 4.69) and IDF (OR = 5.64) metabolic syndrome definitions ($P < 0.05$). The association between VAT and ASAT partial and total volumes with the NCEP and IDF metabolic syndrome is shown in Fig. 2. Independent of measurement site or metabolic syndrome definition, VAT and ASAT were significantly associated with metabolic syndrome ($P < 0.05$); however, the ORs between VAT and metabolic syndrome were higher than for ASAT. The OR for the NCEP metabolic syndrome was higher for total VAT volume (OR = 7.26) and for measures at T12–L1 (OR = 7.46) and L1–L2 (OR = 8.77) than for measures at L4–L5 (OR = 3.94). Similarly, the OR for the IDF metabolic syndrome was higher for total VAT volume (OR = 4.95) and for measures at T12–L1 (OR = 7.08) and L1–L2 (OR = 5.54) than for measures at L4–L5 (OR = 2.99). The association between the NCEP and IDF metabolic syndrome and ASAT

was not altered by measurement site (OR ~ 2.6 , $P < 0.05$).

In a subsample of men with a BMI ≥ 27 kg/m², the magnitude of the associations among waist circumference (NCEP OR = 3.66, IDF OR = 3.48), VAT and ASAT (Fig. 2), and metabolic syndrome was diminished using either metabolic syndrome definition. In this subsample, VAT was significantly associated with NCEP metabolic syndrome at all measurement sites and was significantly associated with IDF metabolic syndrome at intervertebral spaces in the upper abdomen within the region of T10–T11 to L2–L3 ($P \leq 0.05$). ASAT was not significantly associated with either metabolic syndrome definition regardless of measurement site ($P > 0.10$).

CONCLUSIONS— The primary finding of this study was that the mea-

surement site for VAT has a substantial influence on the magnitude of the ORs obtained for metabolic syndrome. Indeed, VAT at the T12–L1 and L1–L2 landmarks had much stronger associations with metabolic syndrome than measures at L4–L5. However, regardless of measurement site and metabolic syndrome criteria, VAT was significantly associated with an increased risk of the presence of metabolic syndrome and, thus, the clinical interpretation was unaltered by measurement protocol.

In this study, regardless of the measurement site and criteria used to identify metabolic syndrome, VAT was more strongly associated with metabolic syndrome than ASAT. These findings extend prior observations reporting that VAT is a stronger predictor of prevalent NCEP metabolic syndrome than ASAT (3,4). Carr et al. (3) measured VAT and ASAT at

Table 2—Relationships between the VAT and ASAT partial volumes at each intervertebral space with the corresponding total VAT and ASAT volume measures

Region of measure (cm ³)	Total VAT Volume (T10–T11 to L5–S1) (l)		Total ASAT volume (T10–T11 to L5–S1) (l)	
	R^2	SEE (%)	R^2	SEE (%)
T10–T11	0.68 ^a	0.85 (27.7) ^a	0.85 ^a	0.52 (14.2) ^a
T11–T12	0.69 ^a	0.82 (26.9) ^a	0.89 ^b	0.45 (12.3) ^b
T12–L1	0.90 ^b	0.47 (15.5) ^b	0.94 ^c	0.34 (9.2) ^{c,d}
L1–L2	0.96 ^c	0.29 (9.6) ^c	0.95 ^d	0.29 (7.8) ^{d,e}
L2–L3	0.96 ^c	0.31 (10.1) ^c	0.96 ^d	0.26 (7.2) ^e
L3–L4	0.91 ^b	0.44 (14.3) ^b	0.94 ^c	0.32 (8.7) ^{c,d}
L4–L5	0.80 ^d	0.65 (21.6) ^d	0.92 ^e	0.38 (10.5) ^{b,c}
L5–S1	0.65 ^a	0.88 (28.8) ^a	0.80 ^a	0.59 (16.1) ^a

All correlations were significant at $P < 0.001$. Significant differences in the correlation coefficients within a given column ($P < 0.05$), as determined using the Hotelling method, are depicted by different superscript letters. Significant differences in SEE within a given column ($P < 0.05$), as determined using the Pitman's test, are depicted by different superscript letters.

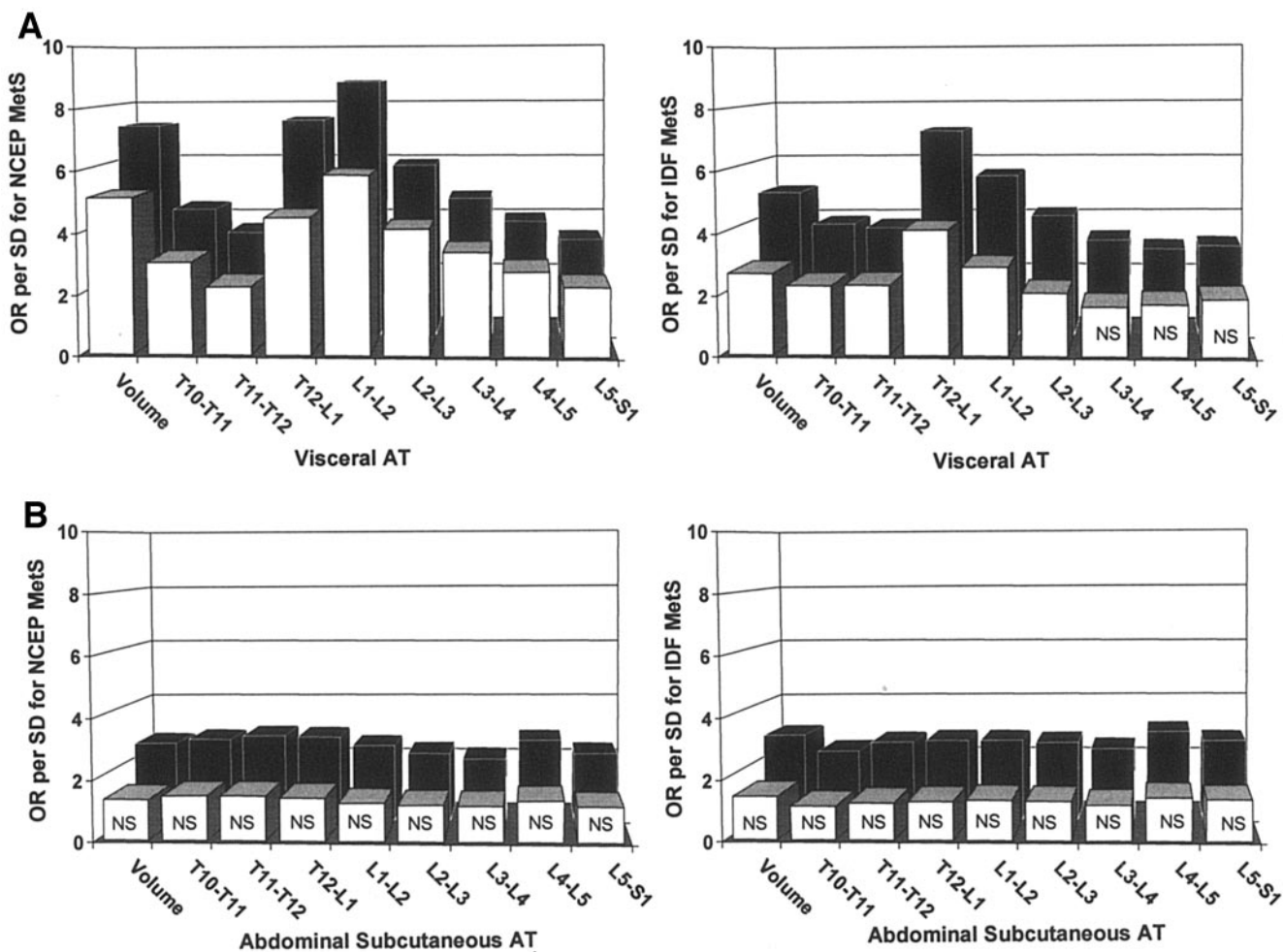


Figure 2—ORs for prevalent metabolic syndrome (MetS) using NCEP and IDF criteria according to measurement location for VAT (A) and ASAT (B) in all men and men with a BMI ≥ 27 kg/m². ■, all men (n = 85); □, men with BMI ≥ 27 kg/m² (n = 40). OR for each measure significant at $P \leq 0.05$ unless otherwise indicated; NS = $P > 0.05$.

the umbilicus, which is proximal to measures at L4–L5. Accordingly, the ORs reported by Carr et al. (3) for VAT (OR = 3.8) and ASAT (OR = 2.9) were similar to those reported in this study at L4–L5 (VAT OR = 3.9, ASAT OR = 2.8). However, the OR we report for metabolic syndrome when VAT was measured at L4–L5 was substantially lower than those observed in the upper abdomen. For example, measurement of VAT at L1–L2 resulted in a twofold higher OR than L4–L5 for NCEP (OR = 8.8 vs. 3.9) and IDF (OR = 5.5 vs. 2.9) metabolic syndrome. Conversely, the association between ASAT for either definition of metabolic syndrome was similar regardless of measurement site (OR = 2.1–3.0).

It is unclear why measurement site would influence the strength of the association between VAT and metabolic syndrome but not ASAT and metabolic syndrome. One reason may be that

whereas the relationship between a single image (e.g., ASAT at a given level) and the total ASAT volume is generally similar throughout the abdomen, we observed marked differences in the association between VAT at a single level and the total VAT volume. If one assumes that the ability to predict VAT or ASAT volume will translate to a corresponding ability to predict metabolic syndrome, then a single image at L1–L2 or L2–L3 may be a better predictor of metabolic syndrome than L4–L5 simply because it is a better predictor of the total adipose tissue volume. Indeed, a single measure at L4–L5 accounted for significantly less variance and had twofold higher error estimates than measures at L1–L2 or L2–L3 ($R^2 = 0.80$ vs. 0.96 and SEE = 22 vs. 10%, respectively). Conversely, the differences in the variance explained and the error estimates for ASAT measured at L4–L5 versus L1–L2 or L2–L3 were subtle ($R^2 =$

0.92 vs. 0.95 and SEE = 10.5 vs. 7.5%, respectively). As such, measurement site may be expected to have a greater influence on VAT- than on ASAT-associated metabolic risk.

A novel finding in this study was that VAT measures at L1–L2 had higher ORs with metabolic syndrome than total VAT volume (Fig. 2). This seems counterintuitive if one assumes that the total VAT mass derived using a multiple image protocol would have a stronger association with metabolic syndrome than one of its component parts (e.g., a single image). This would only be true if one further assumed that adipocyte metabolism throughout the VAT depot was homogeneous. However, it is well established that visceral adipocytes in the omental and mesenteric depots are portally drained and are more sensitive to catecholamine-stimulated lipolysis by comparison to the nonportally drained retroperitoneal adipocytes (8).

Similarly, it is reported that changes in plasma adiponectin levels are largely accounted for by changes in the secretion from omental adipocytes (9). These observations are relevant to our finding as it is reported that the greatest deposition of omental and mesenteric fat is located in the upper abdomen within the region between L1–L2 and L3–L4 (7). Thus, VAT in the upper abdomen might be expected to have a stronger association with hepatic metabolism and associated metabolic derangements than VAT in the lower abdomen. This observation has important implications as it suggests that the measurement of total VAT volume (mass) may not always represent the gold standard when assessing associations between VAT, morbidity, and mortality.

We examined whether the criteria used to identify metabolic syndrome would influence the association with VAT and ASAT. The most clinically relevant metabolic syndrome criteria were proposed by the NCEP in 2001 (11). The recently proposed IDF consensus (12) builds upon the NCEP criteria, but differs in two key aspects. First, the IDF lowered the threshold for waist circumference (from 102 to 94 cm) and fasting glucose (from 6.1 to 5.6 mmol/l). Second, waist circumference is a required component of metabolic syndrome under the IDF criteria, rather than an optional component as used with the NCEP criteria. Given the lower values for waist circumference and glucose, it is not surprising that the IDF metabolic syndrome criteria classify a larger population as having metabolic syndrome than NCEP (16.5 vs. 28.2%, respectively). Nevertheless, although the ORs for IDF metabolic syndrome were consistently lower than those associated with the NCEP definition, we observed a similar influence of measurement site wherein VAT measures in the upper abdomen are more strongly associated with either metabolic syndrome definition. A statistical comparison of the ORs for IDF and NCEP was not possible because the individuals without metabolic syndrome (e.g., the referent groups) differed between the metabolic syndrome definitions. However, for any given study sample, it is expected that the ORs between VAT and IDF metabolic syndrome would be lower than those between VAT and NCEP metabolic syndrome. This is true because many of the additional individuals identified using the IDF criteria have a lower waist circumference and, hence, lower VAT resulting in a smaller

difference in VAT between those with and without metabolic syndrome (NCEP 74 cm² [58%], IDF 59 cm² [48%] at L4–L5). As the ORs reflect the ability of VAT to differentiate those with and without metabolic syndrome, the smaller difference in VAT between those with and without metabolic syndrome weakens the association for IDF metabolic syndrome as compared with NCEP metabolic syndrome.

It is also important to realize that, although the ORs associated with VAT are influenced by the metabolic syndrome definition, the association between VAT and the individual risk factors remain unchanged. In other words, that VAT is associated with a more negative metabolic profile is not altered by the metabolic syndrome definition employed and thus remains an important predictor of metabolic risk. Further, it is important to consider that neither metabolic syndrome criteria nor measurement site altered the statistical significance of the association between VAT or ASAT and metabolic syndrome, and thus the clinical observation was unaltered.

The strengths and limitations of this study include the use of a relatively small opportunistic sample of men who had acquired abdominal CT images at the Cooper Institute. The men in this study are predominantly white and from a middle-to upper-class population. This limits the generalizability of the results of our study but should not affect the internal validity. Indeed, the homogeneity of our study group on socioeconomic factors is a benefit because it reduces the likelihood of confounding by these factors. Future studies should examine whether these observations hold true in women and other racial populations as there are clear sex and racial differences in fat distribution (19) and adipocyte metabolism *in vitro* (20). Further, studies should investigate whether this pattern of association remains true with abdominal adipose tissue changes over time or whether the measurement site influences the relationship between changes in abdominal adipose tissue and obesity-related metabolic risk factors to determine the optimal landmark for the quantification of abdominal adiposity.

In summary, this study is the first to provide evidence that VAT has a stronger association with metabolic syndrome than ASAT independent of measurement site and metabolic syndrome criteria. Although the strength of the association was affected by the measurement site, VAT re-

mained significantly associated with metabolic syndrome regardless of measurement methodology. Thus, the clinical observation and conclusion remains the same regardless of measurement site. This reinforces the notion that elevated visceral adiposity is associated with increased risk of incident metabolic syndrome as characterized by NCEP and IDF and that VAT should be a primary target for pharmacological and lifestyle-based intervention.

Acknowledgments— This research was supported in part by grants from the National Institutes of Health to S.B. (AG06945) and M.L. (HL62508) and from the Canadian Institutes of Health Research to R.R. (MT13448).

The authors thank Elisa Priest and Michael LaMonte for their work related to the CT data management.

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