HHS Public Access

Author manuscript

Am J Physiol Endocrinol Metab. Author manuscript; available in PMC 2017 November 14.

Published in final edited form as:

Am J Physiol Endocrinol Metab. 2007 July ; 293(1): E435–E442. doi:10.1152/ajpendo.00394.2006.

Adipose tissue distribution in relation to insulin resistance in type 2 diabetes mellitus

Koichiro Azuma[1], **Leonie K. Heilbronn**[2], **Jeanine B. Albu**[3], **Steven R. Smith**[2], **Eric Ravussin**[2], **David E. Kelley**[1], and **the Look AHEAD Adipose Research Group**

[1]Department of Medicine, University of Pittsburgh School of Medicine, Pittsburgh, Pennsylvania

[2]Pennington Biomedical Research Center, Louisiana State University, Baton Rouge, Louisiana

[3]Department of Medicine, St. Luke's-Roosevelt Hospital Center, Columbia University, New York, New York

Abstract

Insulin resistance (IR) is typically more severe in obese individuals with type 2 diabetes (T2DM) than in similarly obese non-diabetics but whether there are group differences in body composition and whether such differences contribute to the more severe IR of T2DM is uncertain. DEXA and regional CT imaging were conducted to assess adipose tissue (AT) distribution and fat content in liver and muscle in 67 participants with T2DM (F39/M28, age 60 ± 7 yr, BMI 34 ± 3 kg/m^2) and in 35 similarly obese, non-DM volunteers (F20/M15, age 55 ± 8 yr, BMI 33 ± 2 kg/m^2). A biopsy of subcutaneous abdominal AT was done to measure adipocyte size. A glucose clamp was performed at an insulin infusion of 80 mU · min^{-1}·m^{-2}. There was more severe IR in T2DM (6.1 ± 2.3 vs. 9.9 ± 3.3 mg· min^{-1} · kg FFM^{-1}; $P < 0.01$). Group comparisons of body composition parameters was performed after adjusting for the effect of age, gender, race, height and total fat mass (FM). T2DM was associated with less leg FM (−1.2 ± 0.4 kg, $P < 0.01$), more trunk FM (+1.1 ± 0.4 kg, $P < 0.05$), greater hepatic fat ($P < 0.05$), and more subfascial adipose tissue around skeletal muscle ($P < 0.05$). There was a significant group × sex interaction for VAT ($P < 0.01$), with greater VAT in women with T2DM ($P < 0.01$). Mean adipocyte size (AS) did not significantly differ across groups, and smaller AS was associated with increased leg FM, whereas larger AS was related to more trunk FM (both $P < 0.05$). Group differences in IR were less after adjusting for group differences in leg FM, trunk FM, and hepatic fat, but these adjustments only partially accounted for the greater severity of IR in T2DM. In summary, T2DM, compared with similarly obese nondiabetic men and women, is associated with less leg FM and greater trunk FM and hepatic fat.

Keywords

adipocytes; obesity; body composition; computed tomography

OBESITY IS A RISK FACTOR for type 2 diabetes (T2DM) and insulin resistance (IR). These associations are influenced by adipose tissue (AT) distribution (28). IR is

Address for reprint requests and other correspondence: D. E. Kelley, 807N Montefiore-UPMC; 3459 Fifth Ave., Pittsburgh, PA 15213.

characteristically more severe in T2DM than in similarly obese nondiabetics, but whether this difference is related to differences in body composition is unclear. The amount of visceral AT (VAT) has consistently been demonstrated to correlate with IR (7, 10, 35), an association that has been found in T2DM (12, 13). However, it remains uncertain whether VAT is increased in men with T2DM compared with similarly obese nondiabetic men (1). Increased content of fat within liver and skeletal muscle is also associated with IR and occurs commonly in T2DM (3, 14, 21, 32, 37). In contrast, gluteal-femoral adiposity has a weak association with IR and may instead counterbalance the influences of abdominal adiposity and mitigate risk for IR (43). Several recent studies indicate a relative reduction of gluteal-femoral adiposity in T2DM and in those at increased risk for developing T2DM (38, 41, 42). These observations suggest that the pattern of AT distribution that poses risk for IR in T2DM might derive from a decrease of AT in "metabolically protective" depots, as well as from more AT in "metabolically adverse" depots (e.g., VAT and hepatic fat).

There may be other characteristics of AT that can be considered, such as a preponderance of large adipocytes. A longitudinal study among Pima Indians indicated that large adipocytes in abdominal subcutaneous AT (SAT) poses risk for the development of T2DM independently of fat mass (FM) (45). Danforth (8) postulated that large adipocytes in subcutaneous AT (SAT) denote a saturation of storage capacity in this depot and a disposition for additional fat storage to occur in VAT, liver, and muscle, thereby exacerbating IR. The current study was undertaken to examine the patterns of AT distribution and adipocyte size in T2DM compared with similarly obese nondiabetic men and women and examine the relation-ships with severity of IR.

METHODS

Research volunteers

The present study was performed as an ancillary project at three of the sixteen participating sites of the Look AHEAD Trial (i.e., at the University of Pittsburgh, the Pennington Biomedical Research Center, and the St. Luke's-Roosevelt Hospital Center). The primary goal of the Look AHEAD Trial is to investigate the effects of a lifestyle intervention of weight loss and physical activity vs. that of diabetes support and education on cardiovascular morbidity and mortality (36). Inclusion and exclusion criteria for Look AHEAD, which include a confirmed diagnosis of T2DM, have been previously described in detail (36). For the current study, it was further required that participants be randomized to the lifestyle intervention arm. Additional inclusion criteria were that the participants have a fasting plasma glucose (FPG) ≤ 180 mg/dl and that their diabetes treatment did not include insulin or thiazolidinediones. The current report concerns the baseline (preintervention) findings of 67 volunteers with T2DM (51 non-Hispanic whites, 14 African Americans, 1 Hispanics, and 1 Native American) and a group of 35 nondiabetic (non-DM) men and women. The non-DM participants were recruited to approximate BMI in the T2DM participants and to be age, sex, and ethnicity matched (29 non-Hispanic whites and 6 African Americans) and were studied on a single occasion. Mean age in the participants with T2DM was 59.6 ± 7.2 yr, which was slightly older than the mean age of the non-DM group, 55.1 ± 8.3 yr ($P < 0.01$). The distribution of volunteers at the three sites was that 27 T2DM (F14/M13) and 15 non-DM

subjects (F10/M5) were studied at Pittsburgh, 22 T2DM (F16/M6) and 6 non-DM subjects (F2/M4) were studied at St Luke's-Roosevelt, and 18 T2DM (F9/M9) and 14 non-DM subjects (F8/M6) were studied at Pennington. Informed written consent was obtained from all participants, and the project was approved by each institution's institutional review board as well as by the Look AHEAD Steering Committee.

Body composition

Weight and height were measured on calibrated scales. FM and fat-free mass (FFM) were measured using dual-energy X-ray absorptiometry (DEXA; Hologic QDR 4500A, Waltham, MA). All DEXA scans were analyzed using QDR for Windows v. 11.1 software to determine FM, FFM, leg FM, leg FFM, and abdominal FM (trunk FM). DEXA images were analyzed by the standard default analysis, in which a computer-based algorithm identified the masses of leg FM and trunk FM. The separation between trunk FM and leg FM was made by two oblique lines that pass through the femoral necks.

Three cross-sectional computed tomography (CT) scans, 1 cm in width, centered respectively on the T_{12}-L_1 disc space, the L_4–L_5 disc space, and at the midthigh were obtained to assess hepatic fat content, abdominal AT distribution, and thigh composition. All CT images were analyzed at the University of Pittsburgh using image analysis software (SliceOmatic; Tomovision, Montreal, QC, Canada). To assess hepatic fat content, CT attenuation [Hounsfield units (HU)] was determined in three regions of interest (ROIs) for liver, each ROI of ~120 mm^2. ROIs for the liver were placed manually to avoid major vessels. On the cross-sectional CT images of abdomen and midthigh, the areas for bone, AT, and skeletal muscle were measured electronically by defining for each tissue a range of CT attenuation values: >200 HU for bone, −30 to −190 HU for AT, and 0 –100 HU for muscle, as previously described (17). On abdominal CT images, to determine the respective areas of VAT and SAT, a separation line was drawn manually using a cursor along abdominal wall musculature in continuity with fascia of the paraspinal muscles. Abdominal SAT was further divided into superficial SAT and deep SAT by manually tracing the circumferential superficial fascia, as previously described (23). On thigh CT images, the fascia lata was identified and used to subdivide midthigh AT into SAT and subfascial AT (16).

Metabolic testing and AT biopsy

Participants were admitted to clinical research facilities on the afternoon preceding metabolic studies and the AT biopsy. DEXA and CT imaging were performed on the day of admission. After a standardized dinner (50% carbohydrate, 30% fat, and 20% protein; 7 kcal/kg), participants were fasted over-night. The next morning, a percutaneous biopsy of superficial abdominal SAT was performed, ~10 cm lateral to the umbilicus, using a 5- or 6-mm (ID) Bergstrom needle with suction. Approximately 500 mg of adipose tissue were obtained. Local anesthesia of a 50 –50% mixture of 2% lidocaine and 0.25% bupivacaine (~5 ml) was given prior to the procedure. Approximately 1 h after completion of the biopsy, intravenous catheters were placed in an antecubital vein for infusion of insulin and glucose and in a vein on the dorsum of the contralateral hand for blood sampling, and a heating pad was applied to "arterialize" venous samples. A primed, continuous infusion of insulin (80 $mU \cdot m^{-2} \cdot min^{-1}$) was given for at least 3 h, with the stipulation that insulin be infused for

at least 1 h after reaching a plasma glucose value of 100 mg/dl in those with T2DM. Plasma glucose was measured every 5 min, and a variable infusion of 20% exogenous glucose was used to maintain plasma glucose concentrations. Three blood samples for determination of plasma insulin and FFA were collected during the 30-min period preceding the start of the insulin infusion and during the last 30 min of the clamp. The mean rate of exogenous glucose infusion (GINF) during steady-state insulin infusion, divided by FFM, was used to assess IR. Glucose was analyzed using a glucose oxidase electrode (Syncron CX7; Beckman, Brea, CA). Insulin was measured using an immunoassay on a DPC 2000 (Diagonostic Products, Los Angeles, CA). FFA was measured using a WAKO kit (Wako Chemicals, Richmond, VA).

Adipocyte size

Adipocyte size (AS) and number were determined using a Coulter counter, as previously described (19). These analyses were performed at the Pennington Biomedical Research Center (PBRC). At the time of biopsy, AT was fixed in a solution containing collidine HCl (0.2M) and osmium tetraoxide (31 mg/ml collidine HCl buffer). Fixed samples were shipped to the PBRC, preceding analyses, where they were diluted with 154 mM NaCl and dissociated over 1 wk by the addition of 10 ml of 8 M urea in 154 mM NaCl. The sample was then filtered through a 250-μm nylon filter into a weighed beaker. The volume was increased to 300 ml with 154 mM NaCl containing 0.1% Triton X-100. The cells were counted on a Multisizer-3 (Beckman Coulter, Fullerton, CA) using a 400-μm aperture (dynamic linear range 12–320 μm) and presented as the geometric mean.

Statistical analysis

All statistical analyses were performed using the SigmaStat 3.0 program (SAS Institute, San Rafael, CA). General linear models were used to perform group comparisons of body composition parameters after adjusting for the potential effects of sex, age, race, site, height, and FM and to examine for associations between variables. Two-way interactions of diabetes status with sex and race in relation to body composition parameters were also tested for significance. A dummy variable for clinical site was included in all models. Since the findings for DEXA parameters, such as leg FFM and FM, and trunk FM may be influenced by the length of these respective areas, height was also included in the models. For categorization by race/ethnicity in this statistical modeling, participants were categorized as African American ($n = 20$) or non-African American ($n = 82$; 80 non-Hispanic white, 1 Hispanic and 1 Native American). A P value of <0.05 was considered statistically significant.

RESULTS

Metabolic characteristics

Fasting plasma glucose and insulin were increased in T2DMs compared with the non-DMs ($P < 0.01$), but during the glucose clamp procedure steady-state plasma glucose and insulin were matched across groups. The rate of G_{INF} needed to maintain euglycemia was ~40% lower in T2DM ($P < 0.01$), indicative of more severe IR. Also, fasting and insulin-suppressed plasma FFA were higher in T2DM ($P < 0.01$; Table 1).

Body composition

Unadjusted mean values for the body composition parameters determined using DEXA and CT imaging are presented in Table 2. Comparing unadjusted mean values, BMI, height [167 ± 9 vs. 171 ± 10 cm, T2DM and non-DM, respectively, nonsignificant (NS)], and weight were similar between T2DM and non-DMs, as were FFM (kg), FM (kg) and the percentage of weight accounted for by FM (%FM). Despite the similarities of these systemic metrics of body composition, several group-related regional differences in body composition were observed. Leg FM was 1.3 kg lower in T2DM ($P < 0.05$). The mean CT value for liver (HU) attenuation was lower in T2DM ($P < 0.05$), indicative of higher hepatic fat content in T2DM (34). There were not, however, group differences in abdominal fat distribution as determined by single-slice CT imaging or in trunk FM determined by DEXA imaging, using unadjusted mean values. In the lower extremity, single-slice cross-sectional CT imaging at the midthigh revealed greater subfascial AT in T2DM ($P < 0.05$) and a lower mean value in T2DM for muscle CT attenuation ($P < 0.05$).

Because it is recognized that the amount and distribution of adipose tissue as well as other aspects of body composition are influenced by sex, age, race, and height, the data were adjusted for these variables, and group comparisons were reexamined. Shown in Table 3 are group differences after these adjustments were made (and for comparison, the differences noted for unadjusted comparisons). There were no significant group differences for FFM, FM, and %FM. The earlier noted group differences for less leg FM in T2DM, more subfascial AT, and a lower liver CT attenuation in T2DM remained significant after adjustment for sex, age, race, sites, height, and FM. Additionally, by using these adjusted data, a significant difference in trunk FM was observed; at the mean of adjusted values, trunk FM was 1.1 kg greater in T2DM ($P < 0.05$). These group differences were then further examined.

Leg FM

A linear regression model were used to further appraise the group difference in leg FM, using variables for diabetes status, gender, age, race, height, and FM. At the mean of adjusted values, men had less leg FM (−2.2 ± 0.8 kg, $P < 0.01$), and there was a significant effect of race, with African Americans having more leg FM (+1.2 ± 0.5 kg, $P < 0.05$). The effect of age within this relatively narrow distribution of age (58 ± 8 yr) was not significantly associated with variance in leg FM. Also, this model revealed that each kiloggram increase in FM was associated with +0.3 kg of leg FM ($P < 0.01$). After adjustment for these variables, the effect of T2DM remained significant. At the mean of adjusted values, T2DM was associated with a −1.2 ± 0.4 kg difference in leg FM ($P < 0.01$). This group difference was highly similar to that observed for unadjusted data (Table 3), further indicating an independent effect of T2DM on leg FM.

Trunk FM

After adjustment for sex, age, race, site, height, and FM, a significant group difference in trunk FM was observed. T2DM was associated with +1.1 ± 0.4 kg at the mean of adjusted values ($P < 0.05$). In this model, sex was significantly associated with trunk FM: women had −2.4 ± 0.8 kg compared with men ($P < 0.01$). African Americans had less trunk FM (−1.5

± 0.5 kg, $P < 0.01$). In this relatively narrow age range, age was not significantly associated with variance in trunk FM.

The same approach was used to assess group differences for abdominal AT distribution, using the data of cross-sectional abdominal CT imaging. After adjustment, there were not significant group differences in VAT, superficial SAT, and deep SAT. However, this regression model did reveal a significant sex and group interaction for VAT ($P < 0.01$). At the mean of adjusted values, women with T2DM had greater VAT than did non-DM women (+54 ± 18 cm^2, $P < 0.01$), whereas men with T2DM had a similar amount of VAT as the non-DM men (−67 ± 36 cm^2, NS). There was not a significant group × sex interaction for superficial SAT and deep SAT or for any of the other body composition parameters measured in this study. Taken together with the findings from DEXA, the CT findings indicate that the group difference of increased trunk FM in women with T2DM is mainly accounted for by greater VAT.

Liver CT attenuation

The linear regression model for assessing a group effect on liver CT attenuation is shown in Table 4. Values for liver CT attenuation were not significantly associated with sex, age, height, or FM but were significantly higher in African Americans. At the mean of adjusted values, liver CT attenuation was +8 ± 4 HU greater in African Americans ($P < 0.05$), a difference indicative of less hepatic fat content. After these adjustments, T2DM remained significantly associated with a lower CT attenuation value in the liver (−7 ± 3 HU, $P < 0.05$), indicative of greater hepatic fat content. This adjusted group difference was identical to the unadjusted group difference indicative of an effect of T2DM that is independent of effects of age, sex, race, and FM. In *model 2*, the association of T2DM with lower CT attenuation value in the liver was not influenced by adjusting for VAT. There was neither a significant group × race nor a group × sex interaction for liver CT attenuation.

Midthigh tissue composition

Cross-sectional CT images were obtained at the midthigh level to examine thigh AT distribution between its two main depots of thigh SAT and thigh subfascial AT. Also, the amount of skeletal muscle and its mean CT attenuation value were examined. As earlier noted, there was a significant group difference in subfascial AT on the basis of unadjusted mean values (Table 3), and this difference remained significant (+3.2 ± 1.6 cm^2, $P < 0.05$) after adjusting for the effects of sex, age, race, sites, height, and FM, as shown in Table 5, *model 1*. There were no significant associations of sex, age, race, and height with subfascial AT. At the mean of adjusted values, a 1-kg increase in FM was associated with a 0.5 + 0.1 cm^2 increase in subfascial AT ($P < 0.01$). In *model 2*, we explored the association of subfascial AT with variance in leg FM, as both depots manifested a group difference but no significant association between these depots was observed. In these models, variance in leg FM was not associated with that for subfascial AT, and similar findings were found if thigh subcutaneous fat was used instead of leg FM.

The statistical difference of lower mean CT attenuation for skeletal muscle noted in T2DM, using unadjusted mean values, did not remain significant in comparing adjusted values. The

attenuation of the group difference for muscle CT attenuation was largely due to a negative correlation with age ($P < 0.01$).

Adipocyte size

Biopsy samples of abdominal SAT were obtained to assess mean adipocyte size and examine whether there are group differences. After adjustment for the effects of sex, age, race, site, height, and FM, there was not a significant association of T2DM with adipocyte size (0.86 ± 0.13 vs. 0.78 ± 0.12 μl, T2DM and non-DM, respectively, NS). The linear regression model for adipocyte size is shown in Table 6. There was an association of adipocyte size with FM; at the mean of adjusted values, a 1-kg increase in FM was associated with a +0.013 ± 0.003 μl increase in adipocyte size ($P < 0.01$). We next examined how this association of FM with adipocyte size was influenced by FM distribution. The addition of leg FM to the model revealed that, opposite to the positive association of increased FM with larger adipocyte size, increases of leg FM were significantly associated with a decrease in the size of abdominal adipocyte size (*model 2*). This suggests that the association of FM with larger adipocyte size was due to an association with trunk FM, as is supported by the findings of *model 3*. In neither *model 2* nor *model 3* was DM status statistically significant as a predictor of adipocyte size.

We next undertook an evaluation of the association of adipocyte size with IR, after adjustment for effects of group, sex, age, race, site, height, and FM. After adjustment for FM, there was a negative correlation of G_{INF} with adipocyte size ($P < 0.05$). At the mean of adjusted values, a 1-μl increase in adipocyte size was associated with a -3.2 ± 1.5 mg· kg^{-1}· min^{-1} decrease in insulin-stimulated G_{INF} ($P < 0.05$); yet this had little effect on the association of T2DM with IR. The association of adipocyte size with G_{INF} became non-significant in a model that adjusted for the effect of leg FM and nonsignificant in a model that adjusted for trunk FM.

Relationship of body composition to IR

We next examined the hypothesis that body composition differences related to T2DM contribute to the greater severity of IR in T2DM (compared with similarly obese non-DM individuals). Linear regression models were examined, as presented in Table 7. *Model 1* compared IR (using the parameter of steady-state rates of exogenous G_{INF} during insulin infusion), after adjustment for the effects of sex, age, race, site, height, and FM. There was a significant effect of race; at the mean of adjusted values for G_{INF}, African Americans had a higher values (less IR, $P < 0.01$), whereas the other variables (apart from T2DM) did not significantly account for variance in IR. Following these adjustments, the association of T2DM with more severe IR remained strongly significant ($P < 0.01$). In *model 2*, which added the parameter of leg FM, at the mean of adjusted values a 1-kg increase in leg FM was associated with a $+0.4 \pm 0.2$ mg· kg^{-1} · min^{-1} FFM increase in G_{INF} ($P < 0.01$), slightly decreasing the association of race and group, although each of these associations remained significant. Models were next examined for each of the other three group differences in body composition noted in the current study (i.e., liver CT attenuation, trunk FM, and thigh subfascial AT; *models 3*, *4*, and *5*, respectively). A positive association between higher liver attenuation (and, hence, lower liver fat content) and G_{INF} ($P < 0.05$) was observed and a

negative association for trunk FM ($P < 0.05$). Subfascial AT was not significantly associated with G_{INF}. The addition of these group differences in body composition decreased modestly the strength of the association of DM status with G_{INF}; however, the association between DM status and IR remained significant. The inclusion of VAT in these models (not shown) did not add further significance.

DISCUSSION

IR is typically severe in T2DM and is exacerbated by obesity (24). Since IR is recognized to be influenced by AT distribution and by an accumulation of lipid within liver and muscle (6, 9, 29), the present study was undertaken to address whether there are differences among these body composition parameters in T2DM compared with similarly obese non-DMs. The secondary aim was to address whether such differences contribute to the greater severity of IR in T2DM. Several differences in body composition are related to T2DM. At comparable FM and after adjustment for the effect of sex, race, age, and height, there was significantly less leg FM in T2DM, greater trunk FM, higher fat content in the liver, and a greater amount of subfascial AT, the depot of AT surrounding and interspersed with skeletal muscle. Considered individually, each of these four findings is consistent with prior observations (1, 6, 7, 11, 15, 16, 21, 33, 38, 41). What is novel with regard to the present study is, first, that all of these parameters (as well as others, such as adipocyte size in abdominal SAT, for which group differences were not found) were ascertained in the same set of participants. This enabled a comprehensive delineation of the effect of T2DM on body composition. Second, and arguably more important, was that insulin action was measured in this cohort of research volunteers by use of the rigorous glucose clamp method, which enabled us to examine the relationship between body composition and IR.

In the current study, IR was 40% more severe in men and women with T2DM. There was a significant relationship between severity of IR and three of the components of body composition found to manifest group differences, namely leg FM, trunk FM, and hepatic fat content. Greater amounts of trunk FM and hepatic fat were associated with more severe IR, affirming a well-recognized relationship of an upper-body pattern of obesity and of hepatic steatosis with IR (1, 11, 21, 37). Interestingly, the group difference in leg FM, that is, the decrease observed in T2DM, was also significantly related to more severe IR, and this association remained significant after adjusting for the effect of increased trunk FM and increased hepatic fat. This is an important novel observation. It provides fresh empirical support for the long-held concept that leg FM mitigates the metabolic risks of obesity and that a decrease in this depot is relevant to the pathophysiology of IR in T2DM.

In recent years, a body of data has begun to emerge that leg FM is reduced in T2DM and in those at increased risk for incident T2DM (38, 39, 41, 42). The current findings are strongly consistent. Across groups, despite similar FM, FM in the lower extremities was found to be significantly less in T2DM. There was the expected effect of sex on leg FM, being larger in women, and we observed that leg FM was significantly greater in African Americans. However, these factors did not account for the group differences in leg FM; unadjusted group differences for leg FM were nearly identical to the difference found after adjusting for sex, race, age, FM, and height. Partly, this indicates that the groups were generally well

matched, as indicated by similar mean height, weight, BMI, FM, and %FM. Additionally, the close similarity between adjusted and unadjusted group differences in leg FM also indicates the independent association of T2DM with lower leg FM. The cellular and physiological mechanisms responsible for less leg FM in T2DM are uncertain and warrant investigation. Interestingly, intervention studies with peroxisome proliferator-activated receptor--γ agents improve insulin sensitivity and increase the mass of leg FM, and such changes appear to correlate with systemic improvement of IR in T2DM (20, 44).

The association of T2DM with greater hepatic fat content was also robust, and the group difference was quite similar for unadjusted and adjusted comparisons, indicating an important independent association of hepatic fat content with T2DM. Hepatic steatosis is recognized to occur commonly in T2DM, more commonly than accounted for by obesity (3, 21), a finding confirmed in the present study as group differences were little influenced by adjusting for FM. Prior studies have convincingly demonstrated that hepatic fat is associated with IR (21, 27, 37). This association was also observed in the present study; yet the group difference in hepatic CT attenuation, although robust, appears to account for only a modest proportion of the large group difference in severity of IR, as shown in Table 7.

A number of prior clinical investigations have found a strong association between abdominal adiposity, for visceral adiposity in particular, and IR (4, 24, 35). In the current study, there was a significant association between trunk FM and IR, and there was a significant group difference of more trunk FM in T2DM. This group difference became evident upon adjusted comparisons, and at the mean of adjusted values T2DM was associated with an ~1-kg greater trunk FM, a difference that contributed to group differences in IR. Across groups, greater trunk FM was associated with larger adipocyte size in abdominal SAT, thus manifesting the opposite pattern of association from what was noted for leg FM. These oppositely directed associations of trunk FM and leg FM suggest a physiologically important counterbalancing role for these two major adipose depots. Stated in another manner, these observations suggest that the equation by which AT distribution modulates the relationship between overall FM and IR derives not only from increased abdominal adiposity but additionally from decreased lower-extremity adiposity.

One of the interesting observations was that there was not a group difference for VAT regardless of the significant group difference for trunk FM. This observation is not unprecedented (1) but does need to be carefully qualified. In the current study, men with T2DM had a large amounts of VAT, nearly two to threefold above a threshold value associated with increased metabolic risk (9), but this was not significantly more VAT than similarly obese non-DM men after adjustment for effects of race, age, height, and FM. The second qualification that needs to be emphasized is our finding of a significant group × sex interaction, wherein women with T2DM had significantly greater VAT than non-DM women after adjustment for age, race, height, and FM. It might be speculated that increased VAT in women with T2DM compared with those without diabetes might reflect differences in hormonal regulation (e.g., estrogens, androgens, or glucocorticoids), as has earlier been postulated (2, 5).

Skeletal muscle lipid content is increased in obesity and in T2DM, as has been shown using tissue biopsy studies (18, 30), imaging with magnetic resonance spectroscopy (32), and by comparison of CT attenuation values, as our group reported a number of years ago (22, 26, 40). Whether there is a group effect of T2DM on muscle lipid content after obesity is taken into account is less certain (18), and the current findings, using CT attenuation value as the metric concerning muscle lipid content, suggests that there is not a strong group difference. When unadjusted group means were compared, there was a difference of lower CT attenuation values in T2DM, but this difference was no longer present after adjustment for the effects of age, a well-described determinant of increased muscle lipid content (15). A group difference that did remain significant after adjustment for such factors was greater subfascial AT in T2DM, and here, as with hepatic fat and leg FM, the group difference was almost identical for unadjusted and adjusted mean values, denoting the independent group effect of T2DM. Subfascial AT, a relatively small depot of AT that is located beneath the fascia surrounding muscle and that is interspersed about muscle has previously been shown to correlate with IR and therefore has a quite different relationship than that of the much larger, adjacent depot of leg FM, although the biological basis for these differences is poorly understood.

Biopsy samples were obtained to measure adipocyte size from the layer of superficial SAT. Large adipocytes in abdominal SAT have long been described in obesity, especially in relation to a pattern of upper-body obesity (25, 31). It has been postulated that a preponderance of large adipocytes in SAT signifies saturation of storage capacity and creates a consequent disposition for fat storage in VAT, liver, and muscle (8). We did not find a significant group difference for mean adipocyte size; however, adipocyte size correlated with FM, and larger mean size of adipocytes was related to more severe IR after adjustment for the effect of FM. Moreover, there were novel observations that adjusted for the effects of age, sex, race, and FM; a larger amount of leg FM was associated with smaller mean size of abdominal adipocytes, whereas more trunk FM was associated with larger adipocyte size. Our interpretation is that a relative inefficiency in storage capacity for fat in leg FM acts to divert fat storage to abdominal AT, leading to large adipocytes and the relative saturation of fat storage capacity in this depot. This partitioning between leg FM and trunk FM may in turn set the stage for IR.

The second main hypothesis of the present study was to examine the extent to which group differences in body composition that were identified contribute to the strong group difference between T2DM and non-DMs in severity of IR. We addressed this hypothesis by using multiple regression analysis, adding the potential effects of leg FM, trunk FM, hepatic fat, and subfascial AT to a model in which adjustments were also made for effects of age, sex, race, height, and FM. As shown in Table 7, adjusting for leg FM, trunk FM, and hepatic fat, but not for subfascial AT, did lessen differences between non-DMs and T2DM for severity of IR. However, a clear group difference in severity of IR did remain, indicating that the more severe IR in T2DM compared with similarly obese non-DMs is only partially explicated by the group differences in body composition.

In summary, the present study solidified the emerging body of data that T2DM is associated with lesser AT distributed in the leg region. Furthermore, leg fat was independently and

positively associated with insulin sensitivity after accounting for the negative influence of trunk fat. The current study also reinforced an association between T2DM and hepatic fat that is independent of total fat and VAT. Our findings support the hypothesis that there are differences in AT distribution in T2DM and that these differences contribute to, but do not solely account for, the greater severity of IR that is commonly observed in T2DM. Further research is warranted to determine whether the statistical associations of leg fat and trunk fat with insulin sensitivity reflect physiological cause-and-effect relationships, and if so, what the potential mechanism(s) may be.

Acknowledgments

We acknowledge the other members of the Look AHEAD Adipose Research Group not included in the writing group, who contributed generously to this research project. We are grateful to the participants of the primary Look AHEAD trial and to the control participants for their enthusiastic willingness to participate in this ancillary study. We also acknowledge the help of Dr. Steve Belle of the University of Pittsburgh for assistance with statistical analysis of the data.

Clinical sites and Group members:

Pennington Biomedical Research Center (also the Coordinating Center): George A. Bray, Donna H. Ryan, Donald Williamson, Frank L. Greenway, Allison Strate, Elizabeth Tucker, Kristi Rau, Brandi Armand, Mandy Shipp, Kim Landry, Jennifer Perault.

St. Luke's Roosevelt Hospital Center: Xavier Pi-Sunyer, Jennifer Patricio, Jennifer Mayer, Stanley Heshka, Carmen Pal, Mary Anne Holowaty, Diane Hirsch.

University of Pittsburgh: Carol A. Kelley, Jacqueline Wesche-Thobaben, Rebecca Danchenko, Jowanda Green.

GRANTS

This study was funded by National Institutes of Health Grant DK-60412, with additional support by U01-DK-056990. This work was also supported by the University of Pittsburgh Obesity and Nutrition Research Center (P3-DK-46204), the University of Pittsburgh General Clinical Research Center (MO1-RR-000056), Pennington Biomedical Research Center Clinical Nutrition Research Unit (P30-DK-072476), and the Columbia University Diabetes and Endocrinology Research Center (P30-DK-63608).

References

1. Abate N, Garg A, Peshock RM, Stray-Gundersen J, Adams-Huet B, Grundy SM. Relationship of generalized and regional adiposity to insulin sensitivity in men with NIDDM. Diabetes. 1996; 45:1684–1693. [PubMed: 8922352]
2. Andersson B, Marin P, Lissner L, Vermeulen A, Bjorntorp P. Testosterone concentrations in women and men with NIDDM. Diabetes Care. 1994; 17:405–411. [PubMed: 8062607]
3. Angulo P. Nonalcoholic fatty liver disease. N Engl J Med. 2002; 346:1221–1231. [PubMed: 11961152]
4. Banerji MA, Lebowitz J, Chaiken RL, Gordon D, Kral JG, Lebovitz HE. Relationship of visceral adipose tissue and glucose disposal is independent of sex in black NIDDM subjects. Am J Physiol Endocrinol Metab. 1997; 273:E425–E432.
5. Bjorntorp P. Metabolic implications of body fat distribution. Diabetes Care. 1991; 14:1132–1143. [PubMed: 1773700]
6. Bjorntorp P. Regional fat distribution—implications for type II diabetes. Int J Obes Relat Metab Disord. 1992; 16(Suppl 4):S19–S27.
7. Cnop M, Landchild MJ, Vidal J, Havel PJ, Knowles NG, Carr DR, Wang F, Hull RL, Boyko EJ, Retzlaff BM, Walden CE, Knopp RH, Kahn SE. The concurrent accumulation of intra-abdominal and subcutaneous fat explains the association between insulin resistance and plasma leptin

concentrations: distinct metabolic effects of two fat compartments. Diabetes. 2002; 51:1005–1015. [PubMed: 11916919]

8. Danforth E Jr. Failure of adipocyte differentiation causes type II diabetes mellitus? Nat Genet. 2000; 26:13.
9. Despres JP, Lemieux S, Lamarche B, Prud'homme D, Moorjani S, Brun LD, Gagne C, Lupien PJ. The insulin resistance-dyslipidemic syndrome: contribution of visceral obesity and therapeutic implications. Int J Obes Relat Metab Disord. 1995; 19(Suppl 1):S76–S86. [PubMed: 7550542]
10. Despres JP, Moorjani S, Lupien PJ, Tremblay A, Nadeau A, Bouchard C. Regional distribution of body fat, plasma lipoproteins, and cardiovascular disease. Arteriosclerosis. 1990; 10:497–511. [PubMed: 2196040]
11. Garg A. Regional adiposity and insulin resistance. J Clin Endocrinol Metab. 2004; 89:4206–4210. [PubMed: 15356007]
12. Gastaldelli A, Miyazaki Y, Pettiti M, Matsuda M, Mahankali S, Santini E, DeFronzo RA, Ferrannini E. Metabolic effects of visceral fat accumulation in type 2 diabetes. J Clin Endocrinol Metab. 2002; 87:5098–5103. [PubMed: 12414878]
13. Gautier JF, Mourier A, de Kerviler E, Tarentola A, Bigard AX, Villette JM, Guezennec CY, Cathelineau G. Evaluation of abdominal fat distribution in noninsulin-dependent diabetes mellitus: relationship to insulin resistance. J Clin Endocrinol Metab. 1998; 83:1306–1311. [PubMed: 9543160]
14. Goodpaster BH, Kelley DE, Thaete FL, He J, Ross R. Skeletal muscle attenuation determined by computed tomography is associated with skeletal muscle lipid content. J Appl Physiol. 2000; 89:104–110. [PubMed: 10904041]
15. Goodpaster BH, Krishnaswami S, Harris TB, Katsiaras A, Kritchevsky SB, Simonsick EM, Nevitt M, Holvoet P, Newman AB. Obesity, regional body fat distribution, and the metabolic syndrome in older men and women. Arch Intern Med. 2005; 165:777–783. [PubMed: 15824297]
16. Goodpaster BH, Thaete FL, Kelley DE. Thigh adipose tissue distribution is associated with insulin resistance in obesity and in type 2 diabetes mellitus. Am J Clin Nutr. 2000; 71:885–892. [PubMed: 10731493]
17. Goodpaster BH, Thaete FL, Simoneau JA, Kelley DE. Subcutaneous abdominal fat and thigh muscle composition predict insulin sensitivity independently of visceral fat. Diabetes. 1997; 46:1579–1585. [PubMed: 9313753]
18. Goodpaster BH, Theriault R, Watkins SC, Kelley DE. Intramuscular lipid content is increased in obesity and decreased by weight loss. Metabolism. 2000; 49:467–472. [PubMed: 10778870]
19. Harris RB, Ramsay TG, Smith SR, Bruch RC. Early and late stimulation of ob mRNA expression in meal-fed and overfed rats. J Clin Invest. 1996; 97:2020–2026. [PubMed: 8621790]
20. Kahn SE, Haffner SM, Heise MA, Herman WH, Holman RR, Jones NP, Kravitz BG, Lachin JM, O'Neill MC, Zinman B, Viberti G. Glycemic durability of rosiglitazone, metformin, or glyburide mono-therapy. N Engl J Med. 2006; 355:2427–2443. [PubMed: 17145742]
21. Kelley DE, McKolanis TM, Hegazi RA, Kuller LH, Kalhan SC. Fatty liver in type 2 diabetes mellitus: relation to regional adiposity, fatty acids, and insulin resistance. Am J Physiol Endocrinol Metab. 2003; 285:E906–E916. [PubMed: 12959938]
22. Kelley DE, Slasky BS, Janosky J. Skeletal muscle density: effects of obesity and non-insulin-dependent diabetes mellitus. Am J Clin Nutr. 1991; 54:509–515. [PubMed: 1877507]
23. Kelley DE, Thaete FL, Troost F, Huwe T, Goodpaster BH. Subdivisions of subcutaneous abdominal adipose tissue and insulin resistance. Am J Physiol Endocrinol Metab. 2000; 278:E941–E948. [PubMed: 10780952]
24. Kelley DE, Williams KV, Price JC, McKolanis TM, Goodpaster BH, Thaete FL. Plasma fatty acids, adiposity, and variance of skeletal muscle insulin resistance in type 2 diabetes mellitus. J Clin Endocrinol Metab. 2001; 86:5412–5419. [PubMed: 11701715]
25. Kissebah AH, Vydelingum N, Murray R, Evans DJ, Hartz AJ, Kalkhoff RK, Adams PW. Relation of body fat distribution to metabolic complications of obesity. J Clin Endocrinol Metab. 1982; 54:254–260. [PubMed: 7033275]

26. Larson-Meyer DE, Smith SR, Heilbronn LK, Kelley DE, Ravussin E, Newcomer BR. Muscle-associated triglyceride measured by compute tomography and magnetic resonance spectroscopy. Obesity (Silver Spring). 2006; 14:73–87. [PubMed: 16493125]
27. Marchesini G, Brizi M, Bianchi G, Tomassetti S, Bugianesi E, Lenzi M, McCullough AJ, Natale S, Forlani G, Melchionda N. Nonalcoholic fatty liver disease: a feature of the metabolic syndrome. Diabetes. 2001; 50:1844–1850. [PubMed: 11473047]
28. Marcus MA, Murphy L, Pi-Sunyer FX, Albu JB. Insulin sensitivity and serum triglyceride level in obese white and black women: relationship to visceral and truncal subcutaneous fat. Metabolism. 1999; 48:194–199. [PubMed: 10024081]
29. McGarry JD. Banting lecture 2001. Dysregulation of fatty acid metabolism in the etiology of type 2 diabetes. Diabetes. 2002; 51:7–18. [PubMed: 11756317]
30. Pan D, Lillioja S, Kriketos A, Milner M, Baur L, Bogardus C, Jenkins A, Storlein L. Skeletal muscle triglyceride levels are inversely related to insulin action. Diabetes. 1997; 46:983–988. [PubMed: 9166669]
31. Permana PA, Nair S, Lee YH, Luczy-Bachman G, Vozarova De Courten B, Tataranni PA. Subcutaneous abdominal preadipocyte differentiation in vitro inversely correlates with central obesity. Am J Physiol Endocrinol Metab. 2004; 286:E958–E962. [PubMed: 14970008]
32. Perseghin G, Scifo P, De Cobelli F, Pagliato E, Battezzati A, Arcelloni C, Vanzulli A, Testolin G, Pozza G, Del Maschio A, Luzi L. Intramyocellular triglyceride content is a determinant of in vivo insulin resistance in humans: a 1H-13C nuclear magnetic resonance spectroscopy assessment in offspring of type 2 diabetic parents. Diabetes. 1999; 48:1600–1606. [PubMed: 10426379]
33. Petersen KF, Dufour S, Befroy D, Lehrke M, Hendler RE, Shulman GI. Reversal of nonalcoholic hepatic steatosis, hepatic insulin resistance, and hyperglycemia by moderate weight reduction in patients with type 2 diabetes. Diabetes. 2005; 54:603–608. [PubMed: 15734833]
34. Ricci C, Longo R, Gioulis E, Bosco M, Pollesello P, Masutti F, Croce LS, Paoletti S, de Bernard B, Tiribelli C, Dalla Palma L. Noninvasive in vivo quantitative assessment of fat content in human liver. J Hepatol. 1997; 27:108–113. [PubMed: 9252082]
35. Ross R, Aru J, Freeman J, Hudson R, Janssen I. Abdominal adiposity and insulin resistance in obese men. Am J Physiol Endocrinol Metab. 2002; 282:E657–E663. [PubMed: 11832370]
36. Ryan DH, Espeland MA, Foster GD, Haffner SM, Hubbard VS, Johnson KC, Kahn SE, Knowler WC, Yanovski SZ. Look AHEAD (Action for Health in Diabetes): design and methods for a clinical trial of weight loss for the prevention of cardiovascular disease in type 2 diabetes. Control Clin Trials. 2003; 24:610–628. [PubMed: 14500058]
37. Ryysy L, Hakkinen AM, Goto T, Vehkavaara S, Westerbacka J, Halavaara J, Yki-Jarvinen H. Hepatic fat content and insulin action on free fatty acids and glucose metabolism rather than insulin absorption are associated with insulin requirements during insulin therapy in type 2 diabetic patients. Diabetes. 2000; 49:749–758. [PubMed: 10905483]
38. Seidell JC, Han TS, Feskens EJ, Lean ME. Narrow hips and broad waist circumferences independently contribute to increased risk of non-insulin-dependent diabetes mellitus. J Intern Med. 1997; 242:401–406. [PubMed: 9408070]
39. Seidell JC, Perusse L, Despres JP, Bouchard C. Waist and hip circumferences have independent and opposite effects on cardiovascular disease risk factors: the Quebec Family Study. Am J Clin Nutr. 2001; 74:315–321. [PubMed: 11522554]
40. Simoneau JA, Colberg SR, Thaete FL, Kelley DE. Skeletal muscle glycolytic and oxidative enzyme capacities are determinants of insulin sensitivity and muscle composition in obese women. FASEB J. 1995; 9:273–278. [PubMed: 7781930]
41. Snijder MB, Dekker JM, Visser M, Bouter LM, Stehouwer CD, Yudkin JS, Heine RJ, Nijpels G, Seidell JC. Trunk fat and leg fat have independent and opposite associations with fasting and postload glucose levels: the Hoorn study. Diabetes Care. 2004; 27:372–377. [PubMed: 14747216]
42. Snijder MB, Zimmet PZ, Visser M, Dekker JM, Seidell JC, Shaw JE. Independent and opposite associations of waist and hip circumferences with diabetes, hypertension and dyslipidemia: the AusDiab Study. Int J Obes Relat Metab Disord. 2004; 28:402–409. [PubMed: 14724659]
43. Vague J. Sexual differentiation, a factor affecting the forms of obesity. Presse Med. 1947; 30:339–340.

44. Virtanen KA, Hallsten K, Parkkola R, Janatuinen T, Lonnqvist F, Viljanen T, Ronnemaa T, Knuuti J, Huupponen R, Lonnroth P, Nuutila P. Differential effects of rosiglitazone and metformin on adipose tissue distribution and glucose uptake in type 2 diabetic subjects. Diabetes. 2003; 52:283–290. [PubMed: 12540598]

45. Weyer C, Foley JE, Bogardus C, Tataranni PA, Pratley RE. Enlarged subcutaneous abdominal adipocyte size, but not obesity itself, predicts type II diabetes independent of insulin resistance. Diabetologia. 2000; 43:1498–1506. [PubMed: 11151758]

Table 1

Metabolic parameters in participants with T2DM and in non-DM after an overnight fast and during insulin infusion

	T2DM (*n* = 67)	Non-DM (*n* = 35)
Fasting		
Plasma glucose, mg/dl	137±27	97±8†
Insulin, μU/ml	13±6	9±3†
FFA, mmol/l	0.69±0.18	0.57±0.16†
Clamp		
Plasma glucose, mg/dl	103±6	106±5
Insulin, μU/ml	152±43	166±48
FFA, mmol/l	0.04±0.05	0.01±0.02†
G_{INF}, mg·min^{-1}·kg FFM^{-1}	6.1±2.3	9.9±3.3†

Values are unadjusted means ± SD. T1DM, type 2 diabetes mellitus; non-DM, nondiabetic; FFA, free fatty acid; FFM, fat-free mass; G_{INF}, glucose infusion. Statistical comparisons were made between type 2 DM and nondiabetics.

*$P < 0.05$,

†$P < 0.01$.

Table 2

Body composition analysis by DEXA and CT imaging

	T2DM (F39/M28)	Non-DM (F20/M15)
Weight, kg	94.6±11.2	95.4±14.8
BMI, kg/m^2	34.0±3.0	32.9±2.3
DEXA		
FFM, kg	60.5±10.4	61.3±14.5
FM, kg	34.0±7.5	34.8±6.5
%FM of weight	36.0±7.0	36.7±7.2
Trunk FM, kg	18.7±4.3	18.2±4.3
Leg FM, kg	10.4±3.5	11.7±2.8*
CT of the abdomen		
VAT, cm^2	278±95	259±120
Superficial SAT, cm^2	187±83	191±62
Deep SAT, cm^2	155±58	161±65
CT of the liver		
Liver attenuation, HU	48±13	55±14†
CT of the midthigh		
SAT area, cm^2	128±55	138±46
Subfascial AT area, cm^2	20.5±8.1	16.7±7.2*
Muscle area, cm^2	259±54	277±78
Muscle attenuation, HU	46±5	48±4*

Values are unadjusted means ± SD. DEXA, dual-energy X-ray absorptiometry; CT, computed tomography; FM, fat mass; AT, adipose tissue; VAT, visceral AT; SAT, subcutaneous AT; HU, Hounsfield units. Statistical comparisons were made between T2DM and non-DM.

$^*P < 0.05$;

$^\dagger P < 0.01$.

Table 3

Unadjusted and adjusted group differences between T2DM and non-DM for body composition

	Unadjusted Differences	Adjusted Differences[1]
Weight, kg	−0.8 ± 2.6	0.6+2.2
BMI, kg/m^2	1.1 ± 0.6	1.4+0.6 *
DEXA		
FFM, kg	−0.8 ± 2.5	1.0 ± 1.3
FM, kg	−0.8 ± 1.5	−0.8 ± 1.5
%FM of weight	−0.7 ± 1.5	−1.4 ± 0.9
Trunk FM, kg	0.5 ± 0.9	1.1 ± 0.4 *
Leg FM, kg	−1.3 ± 0.7 *	−1.2 ± 0.4 †
CT of the abdomen		
VAT, cm^2	19 ± 22	14 ± 19
Superficial SAT, cm^2	−4 ± 16	−1 ± 10
Deep SAT, cm^2	−6±13	−3±11
CT of the liver		
Liver attenuation, HU	−7 ± 3	−7 ± 3 *
CT of the midthigh		
SAT area, cm^2	−9 ± 11	−11 ± 7
Subfascial AT area, cm^2	3.8 ± 1.6 *	3.2 ± 1.6 *
Muscle area, cm^2	−18 ± 13	−2 ± 7
Muscle attenuation, HU	−2 ± 1 *	−1 ± 1

Coefficients ± SE and *P* values are shown.

* $P < 0.05$;

† $P < 0.01$. A negative value for group differences indicates a lower value in T2DM.

[1] Data were adjusted for effects of sex, age, race, site, height, and FM, except for the data of weight, BMI, FM, FFM, and %FM, which were adjusted for sex, age, race, and site.

Table 4

Multiple linear regression model for dependent variable liver CT attenuation (HU); regression coefficients (±SE)

	Model 1	Model 2
Diabetes status	−7±3*	−7±3*
Sex	−6±6	−5±7
Age, yr	0.1±0.2	0.1±0.2
Race	8±4*	8±4*
Height, cm	0.4±0.3	0.4±0.3
FM, kg	−0.3±0.3	−0.3±0.3
VAT, cm^2		−0.01±0.02
R^2	0.15	0.15

*$P < 0.05$,

†$P < 0.01$. For DM status, a negative value indicates a lower HU value in T2DM; for sex, a negative value indicates lower HU in men; for race, the positive values indicate higher liver HU in African Americans.

Table 5

Multiple linear regression model for dependent variable thigh subfascial AT (cm^2); regression coefficients (±SE)

	Model 1	Model 2	Model 3
Diabetes status	3.2±1.6*	2.8±1.6	3.0±1.6
Sex	4.3±2.9	3.9±3.0	3.7±3.0
Age, yr	−0.1±0.1	−0.0±0.1	−0.1±0.1
Race	−1.3±1.7	−1.3±1.8	−1.1±1.8
Height, cm	−0.3±0.1	−0.3±0.1*	−0.3±0.1*
FM, kg	0.5±0.1†	0.6±0.2†	0.5±0.1†
Leg FM, kg		−0.3±0.4	
Thigh SAT, cm^2			−0.02±0.02
R^2	0.35	0.35	0.35

*$P < 0.05$;

†$P < 0.01$. For DM status, positive value indicates more subfascial AT in T2DM; for sex, positive value indicates more subfascial AT in men; for race, negative value indicates less subfascial AT in African Americans.

Table 6

Multiple regression model for dependent variable abdominal mean adipocyte size (μl); regression coefficients (±SE)

	Model 1	Model 2	Model 3
Diabetes status	0.07±0.05	0.02±0.05	0.03±0.05
Sex	0.01±0.08	−0.08±0.08	−0.07±0.08
Age, yr	0.00±0.00	−0.00±0.00	0.00±0.0
Race	−0.02±0.05	−0.01±0.05	−0.01±0.05
Height, cm	0.008±0.004 *	0.008±0.004 *	0.008±0.004
FM, kg	0.013±0.003 †	0.023±0.004 †	−0.00±0.01
Leg FM, kg		−0.03±0.01 †	
Trunk FM, kg			−0.03±0.01 †
R^2	0.34	0.44	0.41

* $P < 0.05$;

† $P < 0.01$. For DM status, positive value indicates larger adipocyte size in T2DM; for sex, negative value indicates smaller adipocyte size in men; for race, negative value indicates smaller adipocyte size in African Americans.

Table 7

Multiple linear regression model for dependent variable GINF (mg · min^{-1} · kg FFM^{-1}); regression coefficients (+SE)

	Model 1	Model 2	Model 3	Model 4	Model 5	Model 6
Diabetes status	−3.8±0.6†	−3.3±0.6†	−3.4±0.6†	−3.3±0.6†	−3.9±0.6†	−3.1±0.6†
Sex	0.3±1.1	1.3±1.1	0.2±1.1	1.3±1.1	0.2±1.1	1.2±1.2
Age, yr	−0.0±0.0	−0.0±0.0	−0.0±0.0	−0.0±0.0	−0.0±0.0	−0.0±0.0
Race	2.7±0.7†	2.2±0.7†	2.2±0.7†	2.1±0.7†	2.7±0.7†	1.7±0.7*
Height, cm	−0.0±0.0	−0.0±0.1	−0.0±0.1	−0.0±0.1	−0.0±0.1	−0.0±0.1
FM, kg	0.0±0.1	−0.1±0.1	0.0±0.1	0.2±0.1*	−0.0±0.1	
Leg FM, kg		0.4±0.2†				0.3±0.1*
Liver, HU			0.05±0.02*			0.04±0.02*
Trunk FM, kg				−0.4± 0.2*		−0.1±0.1
Subfascial AT, cm^2					0.0±0.0	0.0±0.0
VAT, cm^2						−0.0±0.0
R^2	0.46	0.50	0.51	0.49	0.46	0.55

*$P < 0.05$,

†$P < 0.01$. Negative value for DM status indicates lower G_{INF} in T2DM; for sex, negative value indicates lower G_{INF} in men; for race, positive value indicates higher G_{INF} in African Americans