Hepatic and Skeletal Muscle Adiposity Are Associated with Diabetes Independent of Visceral Adiposity in Nonobese African-Caribbean Men

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Abstract

Background: Adipose tissue (AT) around and within non-AT organs (i.e., ectopic adiposity) is emerging as a strong risk factor for type 2 diabetes (T2D). Not known is whether major ectopic adiposity depots, such as hepatic, skeletal muscle, and pericardial adiposity (PAT), are associated with T2D independent of visceral adiposity (VAT). More data are particularly needed among high-risk nonobese minority populations, as the race/ethnic gap in T2D risk is greatest among nonobese.

Methods: Thus, we measured several ectopic adiposity depots by computed tomography in 718 (mean age = 64 years) African-Caribbean men on the Island of Tobago overall, and stratified by obesity (obese N = 187 and nonobese N = 532).

Results: In age, lifestyle risk factors, health status, lipid-lowering medication intake, body mass index and all other adiposity-adjusted regression analyses, and hepatic and skeletal muscle adiposity were associated with T2D among nonobese men only (all $P < 0.05$), despite no association between VAT and PAT and T2D.

Conclusions: Our results support the “ectopic fat syndrome” theory in the pathogenesis of T2D among nonobese African-Caribbean men. Longitudinal studies are needed to clarify the independent role of ectopic adiposity in T2D, and to identify possible biological mechanisms underlying this relationship, particularly in high-risk African ancestry and other nonwhite populations.

Keywords: ectopic fat, African ancestry, type 2 diabetes, visceral fat, liver attenuation, skeletal muscle attenuation, myosteatosis

Introduction

OBESITY IS A MAJOR DRIVER of insulin resistance and type 2 diabetes (T2D) risk.¹ However, not all individuals with T2D are obese and vice versa.^{2–4} Specific adipose tissue (AT) patterns and the location of excess adiposity, rather than the total amount of adiposity, may also be important for T2D risk.^{2–5} Moreover, the AT within non-ATs (i.e., ectopic adiposity), composed primarily of white AT, may play an additional role in the development of T2D.^{6,7} Excess visceral,⁸ pericardial,^{9,10} hepatic,^{9,11} and skeletal muscle^{12,13} adiposity are each individually associated with T2D, independent of general obesity. However, the degree of association between each of these adiposity depots and T2D varies by depot type,^{14,15} suggesting that the potential

adverse contribution of ectopic adiposity on T2D is not the same. Few studies have assessed the strength and independence of simultaneously-measured ectopic adiposity depots in T2D risk.

Recent evidence suggests that the relationship between body weight and T2D risk differs by race/ethnicity, with nonwhite populations being highly susceptible to T2D at normal levels of general adiposity as measured by body mass index (BMI).¹⁶ For example, it has been well known that Asians, in particular South Asians, manifest T2D at lower BMI levels compared with whites, due to, in part, race/ethnic differences in visceral adiposity (VAT).^{17,18} African-ancestry populations in the United States and other geographic regions are also disproportionately affected by T2D, but generalized obesity and other risk factors do not

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appear to explain this high T2D burden in this population, and it is still unknown why African-ancestry individuals develop T2D at lower BMI levels compared with whites.^{19–23} African-ancestry individuals have less visceral,²⁴ hepatic,²⁵ and pericardial,²⁶ but more skeletal muscle^{27,28} adiposity compared to European-ancestry individuals.

These differences in ectopic adiposity depots suggest that skeletal muscle adiposity may be a particularly important depot contributing to T2D among African-ancestry individuals. However, comprehensive data on ectopic adiposity in African-ancestry populations are sparse. It remains unclear if hepatic, pericardial, and skeletal muscle ectopic adiposity depots are related to T2D after accounting for VAT. Thus, we examined, for the first time, four simultaneously measured ectopic adiposity depots and tested for their individual and joint association with T2D in a large population-based epidemiologic study among 718 African-Caribbean men.

Materials and Methods

Tobago Health Study

All men in this analysis were from the Tobago Health Study (THS), a population-based, prospective cohort study of community-dwelling men 40 years of age and older, residing on the Caribbean island of Tobago.²⁹ Participants in the THS were recruited without regard to health status and men were eligible if they were ambulatory, not terminally ill, and without a bilateral hip replacement. Men from Tobago are of homogeneous African ancestry with low European admixture (<6%).³⁰ The 765 men for this analysis were randomly recruited for an ancillary study on ectopic adiposity and diabetes, which was completed between 2014 and 2016. All men completed chest, abdomen, and calf computed tomography (CT) scans and interviewer-administered health history questionnaires. Written informed consent was obtained from each participant.

General and ectopic adiposity assessment

Adiposity was measured using standard anthropometry (body weight, BMI, and waist circumference), chest CT (pericardial adiposity-PAT), abdominal CT (VAT, and liver attenuation), and peripheral quantitative CT (calf skeletal muscle attenuation). Body weight was measured to the nearest 0.1 kg on a balance beam scale and standing height was measured to the nearest 0.1 cm using a wall-mounted stadiometer, both without participants wearing shoes. BMI was calculated as body weight in kilograms divided by standing height in meters squared. Waist circumference was measured at the level of the umbilicus or greatest circumference using a flexible tape measure.

CT images of the chest, abdomen, and liver were acquired by a single trained radiologic technician using dual-slice, high-speed NX/I scanner (GE Medical Systems, Waukesha, WI) and then electronically transmitted to the central CT reading center at Vanderbilt University Medical Center, where image analysis and quality control were performed using methods previously described.^{31,32} Briefly, images were analyzed using a dedicated imaging processing workstation with custom-programmed subroutines (OsiriX; Pixmeo, Geneva, Switzerland) and a dedicated pen computing display (Cintiq; Wacom Technology Corporation, Vancou-

ver, WA). A trained analyst manually traced anatomical boundaries (skin, muscular fascia, muscle, and peritoneum) in CT scans. Tissue attenuation thresholds of –190 to –30 Hounsfield Units (HU) were used to distinguish AT voxels in these defined regions. Slices within 15 mm above and 30 mm below the superior extent of the left main coronary artery were selected for PAT measurement. The volume of PAT was determined by summing the AT containing pixels and accounting for the slice thickness. Abdominal VAT was measured from CT scans of 3×3 mm contiguous slices between L4 and L5. VAT was defined as AT located within the peritoneal cavity. Liver attenuation was assessed in HU from three contiguous scan slices taken in the T12 to L1 space. Liver attenuation is well and inversely correlated with hepatic steatosis measured by biopsy.^{33,34} As the amount of hepatic AT increases, the measured liver attenuation decreases (low LA = greater hepatic AT infiltration).³³ Fatty liver disease is defined as liver attenuation ≤40 HU.³⁴

Calf skeletal muscle adiposity was measured at 66% of the calf length, proximal to the terminal end of the tibia, since it has the largest circumference and the lowest variability in composition between individuals.³⁵ Skeletal muscle attenuation reflects intramuscular adiposity content (lower attenuation = greater AT).³⁶ Skeletal muscle attenuation is inversely correlated with skeletal muscle fat content of biopsy specimens.³⁶ Calf skeletal muscle adiposity was measured by peripheral quantitative CT performed with a Stratec XCT-2000 scanner (Orthometrix, Inc.; White Plains, NY) as described previously.¹² All peripheral quantitative CT images were analyzed with STRATEC analysis software version 5.5D (Orthometrix, Inc.) and performed by a trained investigator.

Intrareader technical error in re-analysis of a 5% oversampling of blinded scans was 0.6% for total abdominal volume, 4.4% for VAT volume, 0.98% for liver attenuation, and 1.1% for calf muscle attenuation, indicating high reproducibility of adiposity measures used in our study.

Other characteristics

Demographic, health history, and anthropomorphic characteristics were assessed by trained staff by interview and clinical examinations. Morning fasting blood was drawn at the time of interview, after a minimum of 8 hr of fasting, and was separated into serum samples and aliquoted and frozen at –80°C on the day of blood draw. Fasting serum glucose was measured using an enzymatic procedure in the Advanced Research and Diagnostic Laboratory at the University of Minnesota. Diabetes was defined as a fasting serum glucose level ≥126 mg/dL and/or current self-reported use of diabetes medication, or an affirmative response to the question, “has a doctor ever told you that you have diabetes?” Smoking status was classified both as either current or not, or ever or not, wherein participants reporting smoking <100 cigarettes in their lifetime were considered never-smokers. Self-reported alcohol consumption is very limited in this cohort and was coded as consuming ≥4 drinks per week (yes/no) to identify individuals with greater than average cohort alcohol intake. Self-reported information on walking for exercise or for transportation was recorded because walking is the predominant form of physical activity on the island of Tobago. We used hours of walking per week as a measure of physical activity for these analyses.

TABLE 1. CHARACTERISTICS OF AFRICAN-CARIBBEAN MEN (TOTAL AND BY TYPE 2 DIABETES)

	Total (N=718)	Nondiabetic (N=551)	T2D (N=167)	Age-adjusted P-value
Age (years)	64.0±8.7	63.5±8.6	65.3±8.8	0.016
Walking for exercise (hr/week)	3.3±4.4	3.5±4.6	2.6±3.6	0.08
Current Smoker	7.5%	7.4%	7.7%	0.73
Drink alcohol (4+ per week)	12.6%	13.6%	9.3%	0.16
Watch TV (14+ hr/week)	49.4%	48.8%	51.4%	0.64
Good/excellent health	94.1%	95.7%	89.0%	0.0012
Obesity	26.0%	23.1%	35.9%	0.0001
T2D	23.3%	—	—	—
Fatty liver disease	4.5%	3.3%	8.3%	0.0018
Lipid-lowering medication use	13.5%	9.1%	27.3%	<0.0001
body weight (kg)	85.1±15.6	83.9±15.4	89.0±15.7	<.0001
BMI (kg/m ²)	27.7±4.6	27.2±4.5	29.1±4.7	<.0001
Waist circumference (cm)	98.4±12.8	96.8±12.3	103.7±13.1	<.0001
VAT (cm ³)	96.0±56.2	88.3±50.9	121.0±64.7	<0.0001
PAT (cm ³)	36.2±22.8	33.5±20.5	45.0±27.3	<0.0001
Liver attenuation (HU)	57.1±8.1	58.7±7.6	53.6±8.8	<0.0001
Skeletal muscle attenuation (mg/cm ³)	71.8±4.3	72.3±4.0	70.5±4.9	<0.0001

BMI, body mass index; PAT, pericardial adiposity; T2D, type 2 diabetes; VAT, visceral adiposity.

Participants also rated their overall health status compared to men their own age. Lipid-lowering medications were defined as gemfibrozil and HMG-CoA reductase inhibitors (statin).

Statistical analysis

Adiposity variables were transformed to approximate a normal distribution if necessary. General characteristics were compared using linear regression adjusted for age. Intercorrelations were computed between each adiposity phenotype using age-adjusted Spearman correlations. We examined multicollinearity by assessing the variance inflation factor (VIF). A VIF greater than 2.5 for logistic regression models was used as an indicator of multicollinearity. BMI and lifestyle-adjusted means of adiposity and attenuation were calculated by age group using least squares means, and the trend over the groups was tested using multiple linear regression. Odds ratios (OR) with a 95% confidence interval were calculated per 1-SD increase in each adiposity depot, and per 1-SD decrease in muscle/liver attenuation using multivariable logistic regression. Separate models were fit for each adiposity depot. Initial analyses were adjusted for age. We added potentially important lifestyle factors (alcohol intake, smoking status, and hours of walking per week) into the models. Finally, analyses were further adjusted for BMI and visceral AT. All analyses were performed using SAS version 9.3 (SAS Institute, Cary, NC.)

Results

Baseline characteristics

In a total sample, 26% of participants were obese and 23.3% had T2D (Table 1). Participants with T2D were older, less likely to report excellent/good health, and more likely to report lipid-lowering medication use and to be obese than nondiabetics (Table 1). BMI, weight, waist, and all studied ectopic adiposity depots were significantly higher in participants with T2D compared to those without T2D, independent of age (Table 1).

Inter-relationship of all adiposity measures used in our analyses

All adiposity measures were significantly correlated with each other, but the strength of these associations varied (Table 2; all $P < 0.0001$). In particular, VAT and PAT were highly correlated with each other and with BMI (r values ranged from 0.63 to 0.71). Hepatic and skeletal muscle adiposity were less strongly correlated with BMI, VAT, and PAT (r ranged from 0.32 to 0.50), and weakly with each other ($r = 0.23$).

Adiposity measures across 10-year age groups

We further examined the mean values of adiposity by 10-year age groups, while adjusting for lifestyle risk factors and BMI (Table 3). VAT and PAT increased with advancing age (all $P < 0.0001$). Skeletal muscle attenuation decreased with advancing age ($P < 0.0001$), indicating that adiposity in the skeletal muscle might be increasing with advancing age among African-Caribbean men. Liver attenuation did not differ across age groups.

TABLE 2. AGE-ADJUSTED INTERCORRELATIONS OF ADIPOSITY MEASURES IN AFRICAN-CARIBBEAN MEN (N=718)

	BMI (kg/m ²)	PAT (cm ³)	VAT (cm ³)	Liver attenuation (HU)
BMI (kg/m ²)	—			
PAT (cm ³)	0.63	—		
VAT (cm ³)	0.70	0.71	—	
Liver attenuation (HU)	-0.50	-0.40	-0.49	—
Skeletal muscle attenuation (mg/cm ³)	-0.39	-0.38	-0.32	0.23

All correlation coefficients were statistically significant ($P \leq 0.0001$).

TABLE 3. ADIPOSITY MEASURES ACROSS 10-YEAR AGE GROUPS IN AFRICAN-CARIBBEAN MEN

	Age group (years)				<i>P</i> value for trend
	50–59 (n = 274)	60–69 (n = 272)	70+ (n = 172)	% Difference between men 50–59 and 70+ years of age	
BMI (kg/m ²)	28.7 (0.27)	27.4 (0.27)	26.3 (0.34)	–8.4	<0.0001
PAT (cm ³)	30.7 (1.1)	35.5 (1.1)	45.5 (1.4)	48.2	<0.0001
VAT (cm ³)	82.8 (2.4)	95.0 (2.40)	117.1 (3.1)	41.4	<0.0001
Liver attenuation (HU)	58.3 (0.32)	57.5 (0.31)	57.6 (0.39)	–1.2	0.12
Skeletal muscle attenuation (mg/cm ³)	73.0 (0.23)	72.5 (0.23)	69.0 (0.29)	–5.5	<0.0001

Values are means (SE) adjusted for BMI and lifestyle (alcohol intake, current smoking, and physical activity).

Independent and joint associations of ectopic adiposity with T2D

In the total sample, BMI and all ectopic adiposity depots were significantly associated with T2D, independent of age, lifestyle, health status, and lipid-lowering medication intake (Table 4, all $P < 0.05$). The strongest association with T2D was observed for VAT, followed by hepatic and PAT, BMI, and then skeletal muscle adiposity. We further tested for associations between each adiposity depot and T2D independent of BMI. Each adiposity depot remained statistically significant after adjusting for BMI, although ORs were attenuated (all $P \leq 0.05$). For example, each SD increase in visceral (57 cm³) and pericardial (23 cm³) adiposity, and each SD decrease in liver (8.1 HU) and skeletal muscle (4.2 mg/cm³) attenuation were associated with 55%, 32%, 43%, and 21% greater odds of T2D, respectively. We next tested if the contributions of pericardial, hepatic, and skeletal muscle adiposity to T2D were independent of VAT. After adjusting for VAT, hepatic and skeletal muscle (both $P < 0.05$), but not PAT, remained significantly associated with T2D. Finally, in the model adjusted for all ectopic adiposity depots, only hepatic adiposity remained significantly associated with T2D. In this model, a 1-SD decrease in liver attenuation was associated with 28% increased odds of T2D independent of all other total or ectopic adiposity measures.

Once we divided our participants by obesity status, in obese men only, there was no association between hepatic

and skeletal muscle adiposity with T2D in models adjusted for BMI or VAT, or after the complete mutual adjustment, while the association between VAT and PAT with T2D remained significant after adjusting for BMI (both $P < 0.05$), although not in the mutually adjusted model (Table 5). In contrast, among nonobese men, the association between hepatic and skeletal muscle adiposity and T2D remained significant, independent of BMI or VAT, as well as in the complete model (all $P < 0.05$, Table 6). For example, in the model adjusted for all ectopic adiposity depots, a 1-SD decrease in liver and muscle attenuation was associated with 42% and 28% increased odds of T2D. Interestingly, among nonobese men, the association between VAT and PAT and T2D was not significant in any of the models adjusting for BMI or adiposity (Table 6).

Odds of prevalent T2D by low versus high hepatic and skeletal muscle adiposity groups among nonobese

To determine if hepatic or skeletal muscle adiposity depots make greater contribution to T2D risk among nonobese men, all participants were categorized into high/low hepatic and muscle adiposity groups. For these analyses, high and low adiposity were based on a median split (Fig. 1). Men with low hepatic and low skeletal muscle adiposity served as the reference group. After adjustment for age, lifestyle

TABLE 4. ASSOCIATION OF ADIPOSITY MEASURES WITH TYPE 2 DIABETES IN AFRICAN-CARIBBEAN MEN (N=718, TOTAL SAMPLE)

Adjustment model	BMI, SD = 4.7 kg/m ²	VAT, SD = 57 cm ³	PAT, SD = 23 cm ³	Liver attenuation*, SD = 8.1 HU	Muscle attenuation*, SD = 4.2 mg/cm ³
Model 1 (age adjusted)	1.62 (1.36–1.92)	1.77 (1.47–2.13)	1.66 (1.38–1.99)	1.70 (1.44–2.01)	1.42 (1.20–1.69)
Model 2 (age, lifestyle, health status, and lipid-lowering medication use)	1.55 (1.29–1.87)	1.76 (1.44–2.14)	1.58 (1.30–1.91)	1.62 (1.36–1.93)	1.36 (1.14–1.64)
Model 2 + BMI	—	1.55 (1.19–2.02)	1.32 (1.04–1.68)	1.43 (1.17–1.75)	1.21 (1.00–1.46)
Model 2 + VAT	—	—	1.23 (0.94–1.61)	1.37 (1.12–1.68)	1.22 (1.01–1.46)
Complete mutual adjustment**	1.01 (0.75–1.37)	1.37 (0.99–1.87)	1.04 (0.78–1.40)	1.28 (1.03–1.60)	1.18 (0.96–1.29)

Odds ratios per 1 SD (95% CI) increase or *decrease in adiposity measure.

**Adjusted for age, lifestyle, health status, lipid-lowering medication use, BMI, VAT, PAT, liver attenuation, and skeletal muscle attenuation.

Bold indicates $P < 0.05$.

CI, confidence interval.

TABLE 5. ASSOCIATION OF ADIPOSITY MEASURES WITH TYPE 2 DIABETES IN OBESE AFRICAN-CARIBBEAN MEN ($N=187$, BODY MASS INDEX ≥ 30 kg/m²)

<i>Adjustment Model</i>	<i>BMI, SD=4.7 kg/m²</i>	<i>VAT, SD=57 cm³</i>	<i>PAT, SD=23 cm³</i>	<i>Liver attenuation*, SD=8.1 HU</i>	<i>Muscle attenuation*, SD=4.2 mg/cm³</i>
Model 1 (age adjusted)	1.42 (0.92–2.18)	1.70 (1.13–2.54)	1.68 (1.09–2.57)	1.43 (1.08–1.89)	1.10 (0.80–1.51)
Model 2 (age, lifestyle, health status, and lipid-lowering medication use)	1.37 (0.86–2.17)	1.85 (1.19–2.87)	1.79 (1.13–2.83)	1.40 (1.05–1.88)	1.07 (0.76–1.50)
Model 2 + BMI	—	1.81 (1.11–2.95)	1.72 (1.06–2.78)	1.36 (1.00–1.86)	1.01 (0.70–1.46)
Model 2 + VAT	—	—	1.45 (0.86–2.43)	1.27 (0.93–1.74)	0.99 (0.69–1.41)
Complete mutual adjustment**	0.92 (0.48–1.75)	1.51 (0.85–2.69)	1.27 (0.73–2.22)	1.18 (0.84–1.67)	0.95 (0.65–1.40)

Odds ratios per 1 SD (95% CI) increase or *decrease in adiposity measure.

**Adjusted for age, lifestyle, health status, lipid-lowering medication use, BMI, VAT, PAT, liver attenuation, and skeletal muscle attenuation.

Bold indicates $P < 0.05$.

T2D among obese men = 32.1%.

factors, health status, lipid-lowering medication use, and VAT, nonobese men with low hepatic, but high muscle adiposity, and nonobese men with both high hepatic and muscle adiposity had an increased odds of T2D (OR = 2.07 and OR = 2.29, respectively, both $P < 0.031$). However, men who had high hepatic, but low skeletal muscle adiposity did not have significantly increased odds of T2D, suggesting that skeletal muscle adiposity in nonobese men might be of more importance for T2D risk than hepatic adiposity.

Discussion

The independent contribution of individual ectopic adiposity depots to T2D remains uncertain, largely because studies have rarely simultaneously measured ectopic adiposity from multiple depots. Moreover, little is known about ectopic adiposity and T2D in African-ancestry populations, who have an increased risk of T2D, even at low levels of total adiposity. This study investigated several major adiposity depots in one of the largest cohorts of African-

ancestry men to date. We tested the independent and joint association of adiposity measures with T2D. We found that hepatic and skeletal muscle adiposity were each associated with T2D, independent of VAT among nonobese men, while in contrast, these adiposity depots were not associated with T2D among obese men. Among nonobese men and independent of VAT, increased risk of T2D was observed in men with high skeletal muscle adiposity, irrespective of the degree of hepatic adiposity, suggesting that skeletal muscle adiposity might be a key adiposity depot for T2D among nonobese African-ancestry men.

In our study, VAT showed the highest correlations with BMI, followed closely by PAT. In contrast, hepatic and skeletal muscle adiposity were more moderately associated with BMI and VAT, and weakly with each other. While some studies find that hepatic adiposity correlates strongly with VAT,^{18,37} others report that hepatic^{38–40} and skeletal muscle adiposity⁴⁰ are only weakly correlated with BMI and VAT, suggesting that their determinants and clinical consequences may differ. Our study extends these observations

TABLE 6. ASSOCIATION OF ADIPOSITY MEASURES WITH TYPE 2 DIABETES IN NONOBESE AFRICAN-CARIBBEAN MEN ($N=531$, BODY MASS INDEX < 30 kg/m²)

<i>Adjustment model</i>	<i>BMI, SD=4.7 kg/m²</i>	<i>VAT, SD=57 cm³</i>	<i>PAT, SD=23 cm³</i>	<i>Liver attenuation*, SD=8.1 HU</i>	<i>Muscle attenuation*, SD=4.2 mg/cm³</i>
Model 1 (age adjusted)	2.24 (1.51–3.33)	1.73 (1.34–2.24)	1.48 (1.17–1.89)	1.75 (1.38–2.24)	1.45 (1.16–1.81)
Model 2 (age, lifestyle, health status, and lipid-lowering medication use)	2.28 (1.50–3.47)	1.71 (1.30–2.24)	1.41 (1.10–1.81)	1.72 (1.33–2.22)	1.44 (1.15–1.82)
Model 2 + BMI	—	1.36 (0.98–1.89)	1.12 (0.83–1.51)	1.55 (1.19–2.03)	1.29 (1.01–1.65)
Model 2 + VAT	—	—	1.18 (0.85–1.64)	1.55 (1.17–2.04)	1.34 (1.06–1.71)
Complete mutual adjustment**	1.62 (0.92–2.86)	1.25 (0.85–1.84)	0.92 (0.64–1.31)	1.42 (1.06–1.91)	1.28 (1.00–1.65)

Odds ratios per 1 SD (95% CI) increase or *decrease in adiposity measure.

**Adjusted for age, lifestyle, health status, lipid-lowering medication use, BMI, VAT, PAT, liver attenuation, and skeletal muscle attenuation.

Bold indicates $P < 0.05$.

T2D among nonobese men = 20.2%.

FIG. 1. Odds of prevalent T2D by low versus high hepatic and skeletal muscle groups among nonobese men. Men were separated into groups by median of liver and skeletal muscle attenuation and categorized as either high or low. Shown are the age, lifestyle factors, health status, lipid-lowering medication use, and visceral adiposity, adjusted OR and 95% confidence intervals for prevalent T2D in comparison to the reference group (men with the lowest hepatic and skeletal muscle adiposity). OR, odds ratio; T2D, type 2 diabetes.

in Caucasians to African-ancestry men. These findings suggest that, while BMI may be a good surrogate measure for VAT and PAT, it seems to be a poor surrogate for hepatic and skeletal muscle adiposity. Our findings also support the hypothesis that the contribution of ectopic adiposity depots in various organs to T2D may differ.

Our results suggest that among African-ancestry men, visceral, pericardial, and skeletal muscle adiposity may increase with advancing age, independent of general obesity. Data on longitudinal changes in ectopic adiposity are sparse. A few studies, mainly among Caucasians, have reported a redistribution of adiposity from the subcutaneous to the visceral depot beginning in late middle age until the ninth decade of life.^{41,42} We and others have previously reported that skeletal muscle adiposity increases with advancing age among European- and African-ancestry men,^{12,43} and even accelerates after the age of 65.¹² Similar to our findings of no differences in hepatic adiposity across age groups, the few existing previous studies on this topic also did not find an age-related increase in hepatic adiposity,^{44,45} although no previous study has examined these effects among African-ancestry individuals. Longitudinal studies are needed to better delineate the effects of aging on multiple ectopic adiposity depots among African ancestry, as well as other populations.

An important finding from our work is that all ectopic adiposity depots were associated with T2D, independent of BMI. After adjusting for VAT, pericardial AT was no longer associated with T2D, indicating that it may not contribute to the risk of diabetes beyond that of central adiposity. The most interesting results were obtained among nonobese men. It appears that hepatic and skeletal muscle adiposity are both associated with T2D, independent of BMI and VAT, among nonobese men, and VAT was not even associated with T2D among these men. This is particularly important to note, given that the gap in T2D risk between African- and European-ancestry men is reported to be greatest at low levels of total adiposity.⁴⁶ In addition, it has most recently been reported that, although nonwhite populations generally have higher risk of T2D than whites at any given BMI, these race/ethnic differences are most striking among nonobese, possibly due to differences in accumulation of ectopic fat.¹⁶ Our findings on the association of nonvisceral ectopic adiposity with T2D among nonobese African-ancestry men provide additional evidence to support the previously postulated hypothesis that ectopic fat might provide a better explanation to metabolic disturbances among normal-weight individuals, known as “metabolically obese, but normal-weight” individuals.^{47,48}

Recent studies have shown that hepatic adiposity might be more closely associated with insulin resistance and T2D

than VAT, and that hepatic adiposity may largely account for the relationship between VAT and glucose and insulin metabolism.^{18,49–53} It is important to emphasize that hepatic adiposity seems to be lower among African Americans than in other race/ethnic groups,²⁵ suggesting that this ectopic adiposity depot may not be as important in African-ancestry populations.^{9,54} Most recently, the Jackson Heart Study reported that both visceral and hepatic adiposity are independently associated with cardiometabolic risk among African Americans, although the associations were generally stronger for visceral than for hepatic adiposity.⁵⁴ Interestingly, a study among Hispanic and African-American adolescents has shown that the impact of hepatic adiposity on β -cell function was much more pronounced in African Americans than in Hispanics.⁵⁵ This observation, along with the results of this study, suggests that even though hepatic adiposity tends to be lower in African-ancestry individuals, its association with T2D may be much more pronounced.

Studies have consistently reported that skeletal muscle adiposity may have an important role in T2D, independent of general adiposity.^{12,56–58} However, it remains unclear if this association is independent of VAT, and some still suggest that skeletal muscle adiposity may be a marker of metabolic dysfunction caused by VAT.⁵⁹ Our findings suggest that among nonobese African-ancestry men, skeletal muscle adiposity is associated with T2D even after accounting for VAT. Similarly, greater abdominal skeletal muscle adiposity was associated with a higher prevalence of T2D after adjustment for VAT in the Coronary Artery Risk Development in Young Adults (CARDIA) study among Caucasians and African Americans.³² Comparisons with other studies are somewhat difficult due to differences in the specific skeletal muscle depots assessed across different studies. The CARDIA study assessed adiposity in abdominal skeletal muscles, which are in very close contact with VAT. In contrast, our study assessed adiposity in leg muscles. Indeed, the effects of skeletal muscle adiposity on metabolic risk factors may depend on the function and morphology of muscle fibers,^{60,61} and the proportion of type I and type II fibers⁶⁰ of various skeletal muscles. We have previously reported that association between myosteatosis and insulin resistance is specific for particular abdominal muscle groups.⁶² In addition, changes in muscle quality with aging may also differ for the lower and upper body, and may appear earlier in specific muscle groups in the lower body.⁶³ Also, our study participants have lower BMI and a lower prevalence of other potential confounders (*i.e.*, medication use, admixture, alcohol intake, and smoking) than the CARDIA participants. Although skeletal muscle adiposity and its associations with metabolic disorders need to be further examined by anatomical location (arm vs. abdomen vs. thigh vs. calf), our study suggests that skeletal muscle adiposity may be an independent contributor to T2D among nonobese African Caribbeans, and potentially more important than hepatic adiposity.

The body's response to excess energy accumulation is not identical; there is significant individual variation in how much and where adiposity is stored, and there is a significant variability in body fat distribution for a given BMI.⁶⁴ Race-/ethnic-based differences in ectopic adiposity may also have implications for risk assessment of T2D.⁶⁴ The effects of each adiposity depot vary according with the site and the extent of accumulation.^{14,15} The mechanisms behind

observed differences in association of various ectopic fat depots with T2D are still unknown. Possible mechanisms include endoplasmic reticulum stress, mitochondrial uncoupling, and oxidative stress, all promoting inflammatory response and cell death.¹⁵ Ectopic adiposity was shown to induce both local effects on key target organs as well as systemic effects, such as on the cardiovascular system.¹⁵ In addition, insulin sensitivity and lipid metabolism may substantially differ between organs, and similarly, the extent to which insulin resistance is present in the distinct organs may vary among individuals, and certain population groups.⁶⁵ These interindividual differences in tissue-specific insulin sensitivity may partly be explained by the location where excess lipids are stored. In particular, hepatic and muscle adiposity are linked to the development of liver-IR and muscle-IR, respectively.⁶⁶ Future studies are needed to identify the precise biological mechanisms underlying the relationship between ectopic adiposity and T2D.

This study has potential limitations. First, our analyses may be subject to the limitations of cross-sectional studies such as cohort effects and biases introduced by selective survivorship. A longitudinal study is needed to delineate the effects of ectopic adiposity depots on T2D, as well as the effects of aging on ectopic adiposity. Second, our findings may not apply to younger men, women, or other race/ethnic groups. Third, ectopic pancreatic adiposity, which might be important in T2D,⁶⁷ was not assessed. Fourth, diet and exercise, possible important determinants of adiposity, were not directly assessed in our study. However, in a subset of our participants ($N=355$), we collected objective measures of physical activity using armband accelerometers (SenseWear Pro) for 4–7 days. We found a moderately high correlation between self-reported hours of walking per week and arm-band recorded total hours of any physical activity per week ($r=0.49$, $P<0.05$). This suggests that the walking variable used in the analysis is a good measure of physical activity available in all participants of this study. Fifth, while self-reported levels of typical alcohol intake per week are very low in these men, we did not collect data on patterns of drinking, especially irregular heavy-drinking occasions or binge drinking, which could have contributed to the observed association between liver adiposity and T2D. Finally, there are likely cultural differences in the degree of Westernization between the sub-Saharan Africans, U.K. Africans, African Caribbeans, and African Americans, as well as between various Caribbean islands, and thus, future studies should include populations of African ancestry living outside of the Caribbean region, as well as on other Caribbean islands with predominantly African Caribbeans.

In conclusion, among nonobese African-Caribbean men, hepatic and skeletal muscle adiposity were each associated with T2D, independent of BMI, VAT, and other potentially relevant risk factors. Our findings support the ectopic adiposity syndrome theory⁶⁸ in the pathogenesis of T2D among nonobese African-ancestry men, and emphasize a potential importance of ectopic adiposity distribution in the metabolic complications among normal-weight individuals.

Acknowledgments

The authors would like to thank all supporting staff from the Tobago Health Study Office and the Calder Hall Medical Clinic, as well as all Tobago Health Study participants.

Author Disclosure Statement

No conflicting financial interests exist.

Funding Information

This research was supported by grant R01-AR049747 (PI: J.M.Z.) from the National Institute of Arthritis and Musculoskeletal and Skin Diseases, and grants R03-DK092348 and R01-DK097084 (PI: I.M.) from the National Institute of Diabetes and Digestive and Kidney Diseases. A.L.K. was supported by grant K01-NL125658 from the National Heart, Lung, and Blood Institute.

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