Central fat accretion and insulin sensitivity: differential relationships in parous and nulliparous women

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Abstract

Background/Objectives—Childbearing is associated with a disproportionate accumulation of visceral fat and an increased risk of metabolic disease. However, it is unknown whether the visceral fat accretion associated with pregnancy modifies a woman's risk for metabolic disease. The purpose of this study was to test whether the association between abdominal fat and insulin sensitivity differs by parity status in healthy overweight women.

Subjects/ Methods—Intra-abdominal adipose tissue (IAAT) via CT, body composition by DXA, insulin sensitivity via intravenous glucose tolerance test and minimal model (S_I), HOMA-IR, and cardiorespiratory fitness (VO_{2max}) were assessed in 212 non-diabetic, premenopausal, overweight non-Hispanic white and African American women.

Results—Nulliparous women (n=98) were younger, had less IAAT and higher VO_{2max}, but similar S_I, HOMA-IR, and leg fat, compared to parous (n=114). In nulliparous women, IAAT was negatively associated with S_I, controlling for age, race, and body fat mass ($r=-0.40$, $p<0.001$), but this relationship was attenuated in parous women ($r=-0.15$, $p=0.16$). In multiple linear regression analysis, leg fat and IAAT were significant predictors of S_I in nulliparous, but not parous women.

Conclusions—Results suggest that greater IAAT in parous women does not lead to greater insulin resistance; rather, transient insulin resistance during pregnancy may encourage intra-abdominal fat accumulation that is metabolically benign. This underscores the need to consider parity when assessing cardiometabolic risk.

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Introduction

Obesity, and particularly visceral or intra-abdominal adipose tissue (IAAT) deposition, is strongly related to metabolic disease. Numerous studies have linked insulin resistance and diabetes with the accumulation of fat in the visceral compartment (1) and in other ectopic sites, such as the skeletal muscle and liver (2, 3). In contrast, leg adipose tissue is positively associated with metabolic health (4).

Abdominal fat accumulation is influenced by parity. Parous women have significantly more intra-abdominal adipose tissue than nulliparous women (5), with each child increasing IAAT by approximately 10 cm^2 after adjusting for physical activity, race, and fat gain (6). These and other studies imply that having children may result in a higher risk of diabetes (7), cardiovascular disease (8), and metabolic syndrome (9), but it remains unknown whether the additional IAAT accumulation that has been observed in pregnancy can explain future insulin resistance.

Thus, this study was conducted to examine the relationships among body fat distribution, insulin sensitivity, and parity status. If the negative association between IAAT accumulation and insulin sensitivity persists in parous versus nulliparous women, then increased abdominal fat may explain, at least in part, the increased risk of diabetes and metabolic syndrome in parous women. This study tests whether the association between fat distribution and insulin sensitivity is similar in parous versus nulliparous non-diabetic, overweight, premenopausal women.

METHODS

The data presented here are baseline measures from a weight loss study; the main outcomes have been published (10–12). Based on previously published data (1), we determined that a sample size of 155 subjects is sufficient to achieve a 90% power level with a significance of 0.05 for a multiple regression model with five predictor variables (SAS Studio 3.5).

Subjects were 203 sedentary (no exercise training during the prior year), overweight (BMI > 27 and < 30 kg/m^2) women between the ages of 20–44 years with no history of diabetes. All were tested after a 4-week weight stabilization period during which the subjects were weighed 3 times/week with food provided during the last 2 weeks. Food was provided (20–22% fat, 20–22% protein, and 56–58% carbohydrate) by the General Clinical Research Center (GCRC) Kitchen. Women were admitted to the GCRC 2 days prior to all testing to ensure that physical activity and diet were standardized. After spending the night in the GCRC, testing was completed in the morning in a fasted state. The study was approved by the University of Alabama at Birmingham Institutional Review Board and informed consent was obtained from all subjects.

Cardiorespiratory fitness

A maximal modified Bruce protocol was used to determine VO$_{2max}$ as previously described (13). Heart rate was measured using a POLAR Vantage XL heart rate monitor (Gays Mills, WI, USA). Oxygen uptake and carbon dioxide production were measured continuously

using a MAX-II metabolic cart (Physiodyne Instrument Corporation, Quogue, NY). Gas analyzers were calibrated with certified gases of known concentrations. Standard criteria for heart rate (heart rate within 10 beats/min of estimated maximum), respiratory exchange ratio (RER above 1.2), and plateauing were used to ensure achievement of VO_{2max}. The coefficient of variation for repeat measures of VO_{2max} is less than 3% in our lab.

Body composition and fat distribution

Total and regional body composition, including total fat mass, percent body fat, leg fat mass, and lean body mass were measured by dual-energy X-ray absorptiometry (Prodigy; Lunar Radiation, Madison, WI). The scans were analyzed with the use of ADULT software, version 1.33 (Lunar Radiation). Intra-abdominal adipose tissue (IAAT) was analyzed by computed tomography scanning (CT) (14, 15) with a HiLight/Advantage Scanner (General Electric, Milwaukee, WI) located in the UAB Department of Radiology. Subjects were scanned in the supine position with arms stretched above their heads. A 5 mm scan at the level of the umbilicus (approximately the L4–L5 intervertebral space) was taken. Scans were analyzed for cross-sectional area (cm^2) of adipose tissue using the density contour program with Hounsfield units for adipose tissue set at −190 to −30. All scans were analyzed by the same individual. The coefficient of variation for repeat cross-section analysis of scans among 40 subjects in our laboratory is <2% (14).

Insulin sensitivity

Insulin sensitivity was assessed on an in-patient basis in the GCRC after an overnight fast with an insulin-modified, frequently-sampled intravenous glucose tolerance test (IVGTT). Flexible intravenous catheters were placed in the antecubital spaces of both arms. Three, 2.0 ml blood samples were taken over a 20-min period for determination of basal glucose and insulin (the average of the values was used for basal "fasting" concentrations). At time "0", glucose (50% dextrose; 11.4 g/m^2) was administered intravenously. Insulin (0.02 U/kg, Humulin, Eli Lilly and Co., Indianapolis) was injected at 20 min post glucose injection. Blood samples (2.0 ml) were then collected at the following times (min) relative to glucose administration: 2, 3, 4, 5, 6, 8, 10, 12, 15, 19, 20, 21, 22, 24, 26, 28, 30, 35, 40, 45, 50, 55, 60, 70, 80, 100, 120, 140, 180.

Analyses were conducted in the Core Laboratory of the University of Alabama at Birmingham General Clinical Research Center and Clinical Nutrition Research Center. Glucose was measured using an Ektachem DT II System (Johnson and Johnson Clinical Diagnostics, Rochester, NY). In the Core laboratory, this analysis has a mean intra-assay CV of 0.61%, and a mean inter-assay CV of 2.56%. Insulin was assayed in duplicate 100 μl aliquots using double-antibody radioimmunoassay (Linco Research Inc., St. Charles, MO, Cat #HI-14K). In the Core laboratory, this assay has a sensitivity of 3.35 μIU/ml, a mean intra-assay CV of 3.49%, and a mean inter-assay CV of 5.57%.

Sera were stored at −85°C until analyzed. Glucose and insulin values were entered into the MINMOD computer program (ver. 3, © Richard N. Bergman) for determination of the insulin sensitivity index (S_I) (16). The acute insulin response to glucose (AIRg) was calculated as the incremental insulin area-under-the-curve from minutes 0–10 following

glucose injection using the trapezoidal method. HOMA was calculated using the following equation: [fasting glucose (mg/dl) * fasting insulin (uIU/ml)]/405.

Statistics

Data were analyzed using SPSS version 20. After testing variables for normality, S_I was log-transformed to normalize the distribution of this variable. ANOVA was employed to detect differences in means from groups created by self-reported parity status. Equality of variances was confirmed using Levene's test. Relationships among fat distribution measures and S_I were tested through partial correlations controlled for age, race, and total body fat mass. Multiple linear regression analysis was used to identify significant predictors of S_I within each group, with IAAT, race, age, leg fat mass, and VO_{2max} as independent variables.

RESULTS

Nulliparous women (n=89) were younger, had less IAAT, and higher cardiorespiratory fitness (VO_{2max}), but showed no difference in S_I, HOMA-IR, or leg fat, when compared to parous women (n=114) (Table 1 and Figure 1).

IAAT was strongly and inversely associated with S_I in nulliparous women, but not in parous women (Figure 2A). These relationships remained robust whether the analyses were controlled for total body fat mass, leg fat mass, and/or VO_{2max}.

Leg fat was positively associated with S_I in both parous and nulliparous women, when controlled for age, race, and total body fat mass. This relationship is illustrated in Figure 2B. Leg fat was inversely associated with IAAT in parous (r= −0.363, p< .001), and tended to be associated in nulliparous (r= −0.203, p=.051), women (not shown).

Multiple linear regression analyses indicated that IAAT, age, leg fat, race, and cardiorespiratory fitness were all independent predictors of S_I in nulliparous women. In parous women, however, race was the only one of these predictors that achieved significance (Table 2).

DISCUSSION

The primary finding of this study was that parity decreases if not eliminates the relationship between fat distribution and insulin sensitivity/resistance. Although IAAT is often thought to "cause" insulin resistance, our data show that insulin sensitivity did not differ with parity status, despite higher IAAT in parous women. Importantly, the expected inverse relationship between IAAT and insulin sensitivity was observed in nulliparous but not parous women. This demonstration of dissociation implies that higher levels of IAAT may not necessarily impact insulin resistance as strongly in parous as in nulliparous women. We also observed that the "protective" effect of leg fat decreased with parity, despite no difference in leg fat or insulin sensitivity/resistance between parity status. Thus, the commonly observed associations among fat distribution and insulin sensitivity/resistance may indicate co-occurring epi-phenomena, or may reflect the ability of insulin sensitivity/resistance to regulate fat patterning.

In our data, IAAT was associated with S_I in nulliparous but not in parous women. The cause of the dissociation of IAAT from S_I after pregnancy is unclear. One theory we propose is that insulin sensitivity leads to peripheral fat accretion, whereas insulin resistance encourages visceral fat accumulation. As women develop transient insulin resistance during pregnancy (17), gestational weight gain leads to deposition of IAAT. After pregnancy, insulin sensitivity returns, however epigenetic programming results in maintenance of IAAT stores.

Leg fat is considered a metabolically benign adipose depot; i.e., it is not associated with adverse health consequences. In fact, clinical research studies (4) and epidemiological observation (18, 19) have both shown that greater leg fat is associated with more favorable health outcomes. Leg fat is characterized by a low free fatty acid flux. Thus it is possible that leg fat protects the system from excessive exposure to free fatty acids, which can promote insulin resistance (20). However, an alternative explanation is that metabolically healthy, insulin sensitive individuals are prone to depositing fat in the leg area. In contrast, because leg fat accrual is regulated by insulin, insulin resistant individuals are less able to deposit fat in the leg area. These relationships support the concept that fat is partitioned based on underlying endocrine factors that direct lipid storage to one depot vs the other. Thus, fat pattern (intra-abdominal vs leg) is a marker for insulin sensitivity/resistance.

When looking at the association between leg fat and insulin sensitivity, greater leg fat was associated, as expected, with higher S_I in nulliparous women. However in parous women, leg fat was not associated with insulin sensitivity. Thus, parity essentially eliminated the association of leg fat with insulin sensitivity. It is important to note that neither leg fat mass nor S_I differed with parity; only the association amongst variables differed. Although we do not know the basis for this dissociation, it is possible that the metabolic properties of leg fat are altered following parity, perhaps due to exposure to the high concentrations of reproductive hormones during pregnancy.

Transient insulin resistance develops over the course of a normal pregnancy. Estrogen, progesterone, and human placental growth hormone (hPGH) (21) increase steadily throughout pregnancy and are thought to be responsible for alterations in both insulin sensitivity (17) and beta cell activity (22). Additionally, increased estrogen levels also have been associated with subcutaneous fat deposition in mice (23). Therefore, increased estrogen levels may explain the reduced association between leg fat deposition and insulin sensitivity in this cohort of women. Moreover, human placental lactogen (hPL) activity is affected by circulating levels of serum glucose and its function, at least in part, is to promote lipolysis (24). This hormone is elevated in response to hypoglycemic states and inhibited in hyperglycemic environments, and therefore may have a contributing role in adipose storage during a state of insulin resistance. How these hormones, and others, influence body fat distribution in obese women during pregnancy is uncertain.

The limitations of this study should be considered. This study is cross-sectional and does not necessarily capture the changes that occur during pregnancy. Additionally, the time span since gestation is inconsistent among these women. It is possible that IAAT accumulation and its relationship with insulin sensitivity change over the course of time since gestation.

The S_I index reflects both insulin-stimulated glucose uptake and insulin inhibition of hepatic glucose production. Since S_I is a "whole body" index, it is difficult to interpret. One consideration is that the relative contributions by muscle and liver may differ among the sub-groups in this study. Furthermore, this study included both African American and non-Hispanic white women; racial differences have been reported among the relationships involving fat distribution and insulin sensitivity/resistance (2). As seen in the regression models, our data showed an influence of race on S_I and therefore may be an important consideration, especially in parous women. While beyond the scope of the current project, racial differences in these relationships should be examined in future studies.

In conclusion, the relationships among fat distribution measures and insulin sensitivity/resistance in women vary as a result of reproductive history. Greater IAAT in parous women was not associated with lower S_I, and the "protective" effect of leg fat was less apparent in parous women. Therefore, if childbearing is indeed associated with a higher risk of cardiometabolic disease, it may not be due to metabolic impairment caused by excess visceral fat. On the contrary, we propose that the increased accretion of visceral fat associated with pregnancy is due to transient insulin resistance, which becomes resolved shortly post-partum. This underscores the need to consider parity when assessing cardiometabolic risk.

Acknowledgments

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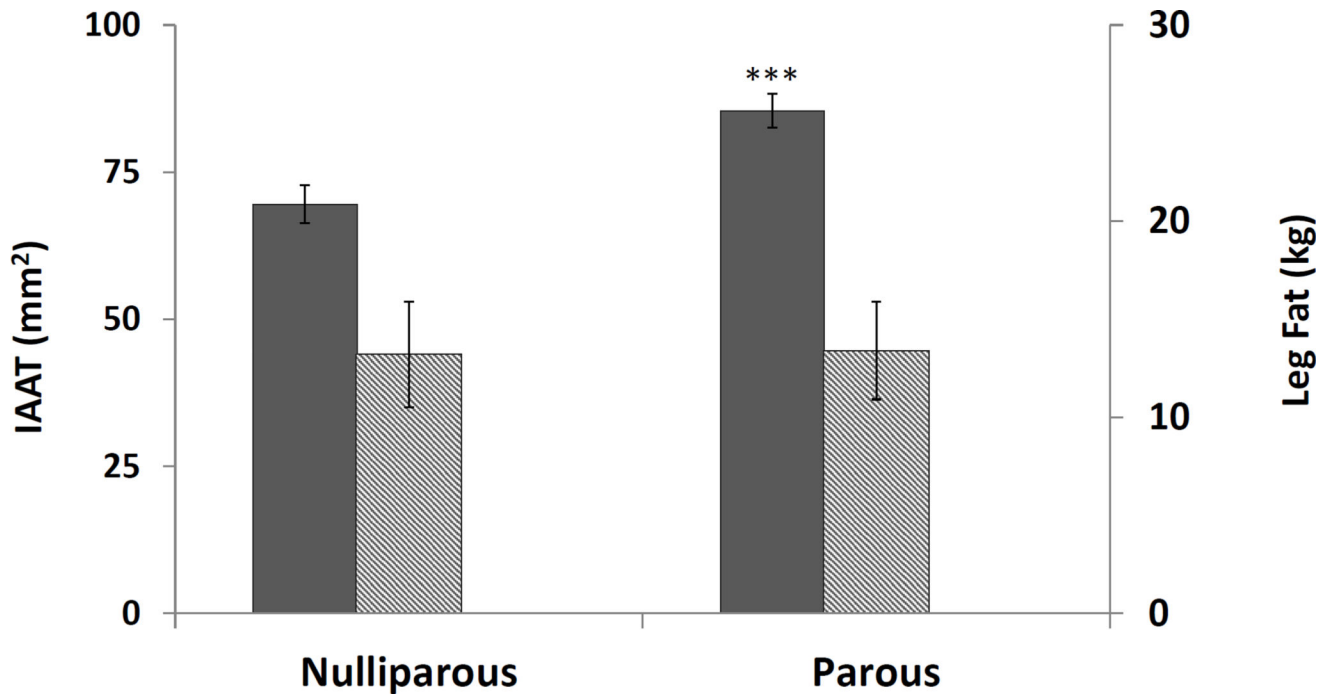
Figure 1.
Abdominal and leg fat accumulation in overweight nulliparous and parous women.
Solid bars represent intra-abdominal adipose tissue (IAAT) in nulliparous (N=98) and parous (N=104) women. Striped bars represent leg fat mass. ***Indicates mean difference between parous and nulliparous women at p<0.001.

A

○ NP: r= -.398***
● Parous: r= -.145

B
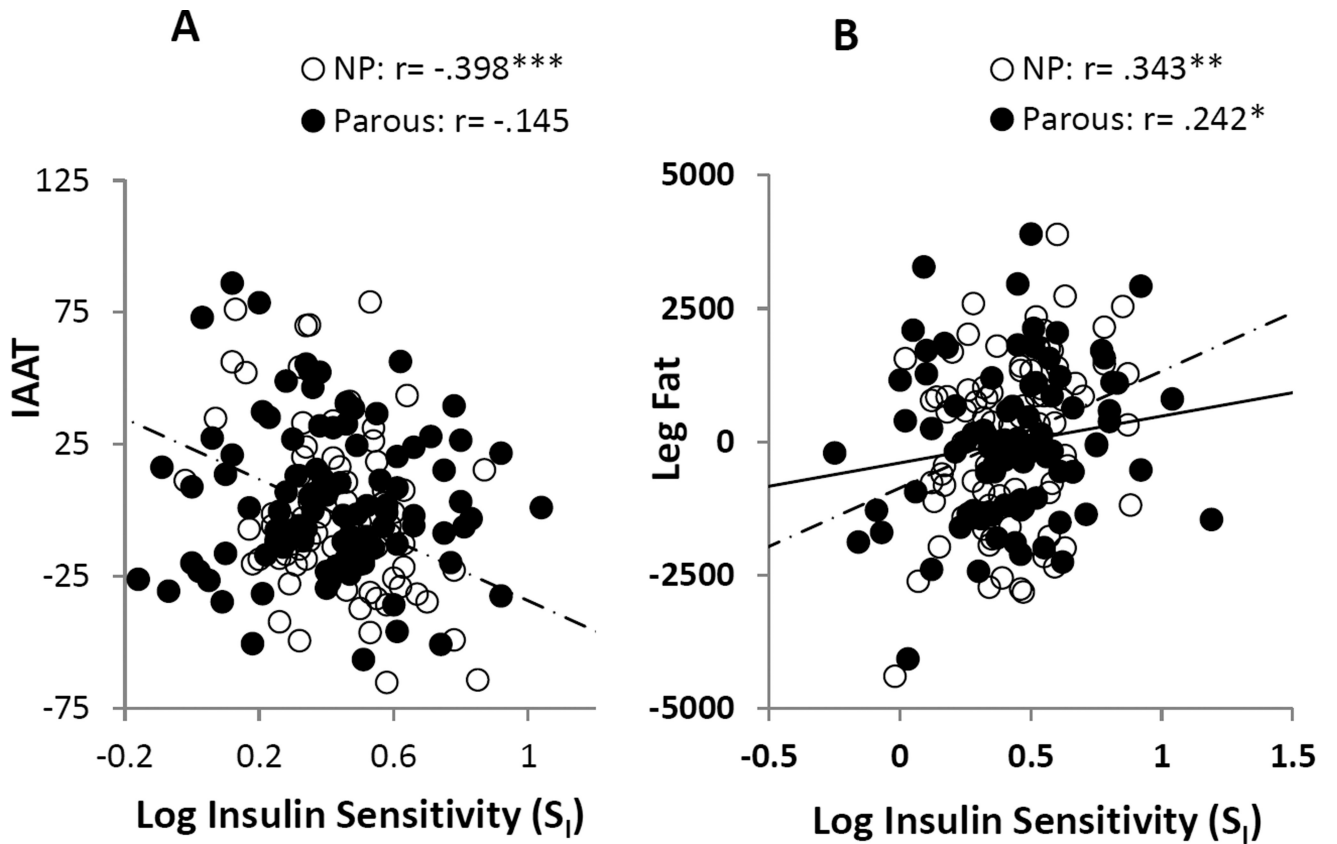
○ NP: r= .343**
● Parous: r= .242*

Figure 2.
A and B. Correlations between intra-abdominal adipose tissue (IAAT) or leg fat (as residuals controlled for age, race, and body fat) and insulin sensitivity index (Log $_{10}$ S_I) in parous vs. nulliparous women. Trend lines are displayed for significant relationships only. Nulliparous women (NP) are indicated with open shapes and significant relationships illustrated with dashed lines. Parous women are indicated with filled shapes and solid lines. Two-tailed level of significance indicated with *p<0.05, **p<0.01, or ***p<0.001.

Table 1

Characteristics of study population by parity status.

	Parous	Nulliparous
N	114	98
Race (AA/ NHW)	58/56	50/48
Age (years)	36.4 ± 5.1	31.3 ± 6.4 [*]
BMI	27.5 ± 4.7	27.3 ± 5.1
# Children	1.8 ± 0.7	0.0 ± 0.0 [*]
Lean Mass (kg)	43.5 ± 4.2	43.5 ± 4.3
$S_I(x10^{-4}min^{-1}(\mu U/ml))$	3.2 ± 2.2	3.0 ± 1.4
HOMA-IR	2.6 ± 0.9	2.4 ± 0.8
VO_{2max} (ml/kg.min)	27.7 ± 3.6	29.5 ± 4.0 [*]
IAAT (cm^2)	85.4 ± 30.3	69.6 ± 31.4 [*]
SAT (cm^2)	333.4 ± 79.3	331.2 ± 99.8
Leg tissue (kg)	27.4 ± 3.5	27.3 ± 3.6
Leg fat (kg)	13.4 ± 2.5	13.2 ± 2.7
% Body Fat	43.6 ± 3.6	43.0 ± 4.2
Glucose (mg/dl)	88.0 ± 6.3	86.7 ± 6.7
Insulin ($\mu U/ml$)	11.7 ± 3.9	11.3 ± 3.4

Data are reported as mean ± standard deviation, unless otherwise indicated.

[*] Indicates mean difference at $p < 0.01$.

AA, African American; NHW, non-Hispanic white; BMI, body mass index; S_I, insulin sensitivity index; HOMA-IR, homeostasis model assessment of insulin resistance; VO_{2max}, maximal oxygen consumption; IAAT, intra-abdominal adipose tissue; SAT, subcutaneous abdominal adipose tissue.

Table 2

Multiple linear regression in all women and grouped by parity status.

Group	Variables	Std Beta	p	Model R^2	F	p
All women	DV: S_I log 10			.237	12.100	<0.001
n=204	IAAT	−.230	.004			
	Age	.138	.039			
	Leg Fat Mass	.217	.001			
	Race	−.446	.000			
	VO_{2max}	.147	.040			
Nulliparous	DV: S_I log 10			.386	10.194	<0.001
n=89	IAAT	−.297	.008			
	Age	.289	.002			
	Leg Fat Mass	.395	.000			
	Race	−.441	.000			
	VO_{2max}	.247	.013			
Parous	DV: S_I log 10			.154	3.375	0.008
n=100	IAAT	−.133	.256			
	Age	−.031	.760			
	Leg Fat Mass	.131	.203			
	Race	−.399	.000			
	VO_{2max}	.069	.515			

S_I, insulin sensitivity index; IAAT, intra-abdominal adipose tissue; VO_{2max}, maximal oxygen consumption.