Resistance to the antilipolytic effect of insulin in adipocytes of African-American compared to Caucasian postmenopausal women

Susan K. Fried,[1,*,†] Thomas Tittelbach,[*] Jacob Blumenthal,[*] Urmila Sreenivasan,[*] Linda Robey,[*] Jamie Yi,[*] Sumbul Khan,[*] Courtney Hollender,[*] Alice S. Ryan,[*,§] and Andrew P. Goldberg[*,§]

Geriatric Research and Education Clinical Center, Baltimore Veterans Administration Medical Center,* Baltimore, MD; and Divisions of Endocrinology, Diabetes and Nutrition,† and Gerontology,§ Department of Medicine, University of Maryland School of Medicine, Baltimore, MD

Abstract **High fatty acid (FA) flux is associated with systemic insulin resistance, and African-American (AA) women tend to be more insulin resistant. We assessed possible depot and race difference in the antilipolytic effect of insulin in adipocytes isolated from abdominal (Abd) and gluteal (Glt) subcutaneous (sc) adipose tissue of overweight, postmenopausal AA and Caucasian (C) women. Percent body fat, fasting insulin, visceral adiposity, and adipocyte size was higher in AA women. Disinhibited lipolysis (presence of adenosine deaminase) per unit adipocyte surface area was similar in Abd and Glt and in AA and C. However, rates of 'basal' [submaximal phenylisopropyl adenosine (PIA)-suppressed] and insulin-suppressed lipolysis were higher in Abd of AA compared with C women even after adjustment for percent fat and visceral fat area. The race difference in rates of PIA- and insulin-suppressed lipolysis in AA were correlated with their hyperinsulinemia, but AA race, independent of fasting insulin, was associated with lower responsiveness (percent suppression) to submaximal insulin concentrations, although sensitivity (ED50) was not affected. Overall, these data are consistent with the hypothesis that decreased responsiveness of Abd adipocytes to antilipolytic effectors may contribute to higher FA availability and thereby to racial differences in insulin resistance.**—Fried, S. K., T. Tittelbach, J. Blumenthal, U. Sreenivasan, L. Robey, J. Yi, S. Khan, C. Hollender, A. S. Ryan, and A. P. Goldberg. **Resistance to the antilipolytic effect of insulin in adipocytes of African-American compared to Caucasian postmenopausal women.** ***J. Lipid Res.*** **2010.** 51: **1193–1200.**

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Increased availability of nonesterified fatty acids (FA) contributes to the metabolic dysfunction associated with obesity, including systemic insulin resistance and hyperlipidemia (1). It is well established that central or upper body obesity, independent of total body fat, is associated with elevated systemic nonesterified FA turnover in premenopausal women. In vivo studies of FA fluxes show that on a per gram basis, abdominal (Abd) subcutaneous (sc) fat is the main source of the high systemic lipolysis in upper body compared with lower body obese premenopausal women and that leg fat is less lipolytically active (1). However, microdialysis studies detect no depot difference in lipolysis in nonobese, premenopausal Abd and femoral sc adipose tissue (2). Nevertheless, both techniques reveal similar responses to the antilipolytic effect of insulin in Abd sc and lower body adipose tissues. In vitro studies also show similar basal lipolytic rates in Abd and gluteal (Glt) or femoral adipocytes of premenopausal women, but Glt adipocytes are more sensitive to insulin (3, 4). Whether this depot difference exists in postmenopausal women is not known, so this was one of the objectives of the current study.

Overweight African-American (AA) women are more hyperinsulinemic and resistant to insulin's glucoregulatory actions than body fat-matched Caucasian (C) women (5, 6). Thus, it is logical to postulate that AAs may also exhibit higher rates of lipolysis and resistance to the antilipolytic effect of insulin. However, results in the literature on this topic are inconsistent. In premenopausal women,

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Abbreviations: AA, African-American; Abd, abdominal; ADA, adenosine deaminase; C, Caucasian; FCW, fat cell weight; Glt, gluteal; HOMA-IR, homeostasis model of insulin resistance; SAT, subcutaneous adipose tissue; sc, subcutaneous; UBO, upper body obese; VAT, visceral adipose tissue; PIA, phenylisopropyladenosine; MANOVA, multivariate analysis of variance.

[1] To whom correspondence should be addressed. Current address: 650 Albany St., EBRC Rm 815, Boston, MA 02118
e-mail: skfried@bu.edu

in vitro studies of Dowling et al. (7) showed that Abd adipocytes from C compared with AA upper body obese (UBO) women (based on waist to hip ratio, UBO) are less sensitive to suppression by adenosine and insulin; however, subsequent studies showed the apparent race effect in UBO women was attributable to the lower visceral adiposity and hyperinsulinemia of the AA women (8). In vivo studies from the same laboratory detected no race difference in rates of baseline or insulin-suppressed FA turnover in premenopausal women with similar levels of visceral adiposity, but resistance to the antilipolytic effect of insulin administration on FA turnover was associated with higher visceral AT accumulation, independent of race (9). In contrast, Racette et al. (10) found lower in vivo FA turnover in premenopausal AA women who were less viscerally obese and had similar insulin levels to the C women. Barakat et al. (11) also reported lower basal lipolytic rates in omental adipose tissue fragments from morbidly obese AA compared with C women. In contrast to these results in obese AA women, higher rates of lipolysis (glycerol release) measured in vivo by microdialysis (12) were detected in South African black women, and Butherlezi et al. (3) found higher circulating FA and lower responsiveness to the ability of insulin to suppress isoproterenol-stimulated lipolysis in isolated adipocytes from both the Abd and femoral depots of the South African black women. However, these women were relatively insulinopenic, which contrasts with the hyperinsulinemia typical of AA women. The current study was designed to determine whether there are race differences in the regulation of lipolysis in Abd and Glt adipocytes of postmenopausal AA and C women. Because racial differences in lipolysis could be associated with race differences in fat distribution and insulinemia, we analyzed the potential confounding influence of these variables.

METHODS

Subjects and characterization

Weight stable (BMI 24–47 kg/m^2) postmenopausal C and AA women between the ages of 47 and 78 years, at least 1 year past menopause (follicle stimulating hormone > 30 IU/ml) were recruited from the Baltimore metropolitan area. Volunteers underwent a 2 h oral glucose tolerance test and those with 2 h glucose > 200 mg/dl or fasting glucose > 126 mg/dl were excluded (6). Subjects taking hormone replacement therapy, statins, and β-blockers (but not other antihypertensives) were excluded from our lipolysis analyses. Fat mass was determined by dual-energy X-ray absorptiometry (Model Prodigy LUNAR GE version 7.53.002). CT scans at L2-L3 and L4-L5 were performed using a Siemens Somatom sensation 64 Scanner. Analyses of visceral and sc fat areas were performed using Medical Image Processing, Analysis and Visualization, v.7.0.0 software. Serum glucose was measured by the glucose oxidase methods (2300 STAT Plus, YSI, Yellow Springs, OH). Immunoreactive insulin was determined by radioimmunoassay (Linco Research Inc., St. Charles, MO). The Institutional Review Board of the University of Maryland approved all methods and procedures, and each participant provided informed written consent.

Adipocyte isolation and lipolysis measurements

Aspirates of the Abd sc and Glt were taken with a cannula under local lidocaine anesthesia as previously described and immediately transported to the laboratory (9). Adipocytes were isolated by collagenase digestion (1 mg/ml) according to Honnor et al. (13) in Kreb-Ringer bicarbonate buffer with 4% BSA and 200 nM adenosine, pH 7.4, under an atmosphere of 95% O_2:5% CO_2. Using the same buffer without collagenase, cells were washed three times by floatation and adipocytes were resuspended at a concentration of 5–7% (v/v). Aliquots of cells (0.5 ml) were added to 13 × 100 mm polyethylene test tubes to which had been added 4 μg/ml adenosine deaminase and other reagents as noted in specific experiments (i.e., varying concentrations of phenylisopropyl adenosine or insulin). Preliminary experiments indicated that lipolysis, as assessed by glycerol release, was linear with time over the 2 h incubation. After 2 h incubation with shaking (60 cycles/min) at 37°C under an atmosphere of 95% O_2:5% CO_2, the reaction was stopped by placing cells in ice bath, and the media below the floating fat cells was removed and frozen at −80 C. Glycerol concentration in the medium was measured fluormetrically in neutralized perchloric acid extracts (14). Nonesterified FA concentration in the medium was measured using a kit from Wako (Richmond, VA).

Fat cell weight (FCW) was measured by the diameters of at least 300 adipocytes using the photomicroscopic method of Lavau et al. (15) and surface area was calculated according to Leibel et al. (16). Only cells >50 μm in diameter were included in the final calculation of mean FCW.

Statistics

Repeated measures ANOVA and paired *t*-tests were used to assess within-subject effects of depot, and repeated measures multivariate (MANOVA) was used to assess the influence of potentially confounding factors on the race difference, such as insulin, glucose, and measures of body fat and body fat distribution. Posthoc independent *t*-tests were used to test group (race) differences using JMP (SAS Software). Variables that were not normally distributed (insulin, glycerol release, ED_{50}) were log-transformed prior to statistical analysis. A least squares model was used to determine the independent effects of race and baseline lipolysis (ADA) on insulin responsiveness (percent suppression). Data are reported as means ± SEM; significance was two-tailed at $P < 0.05$.

RESULTS

Subject characteristics

The AA and C postmenopausal women studied were similar in age, but BMI and percent fat were significantly higher in the AAs (**Table 1**). There was no race difference in waist circumference or trunk fat by dual-energy X-ray absorptiometry, but leg fat was greater in AA. The relative distribution of fat in the trunk and leg (ratio to total body fat) and waist-hip ratio did not differ significantly between the races. Visceral fat area (at L2-3 or L4-5) was similar in C and AA women, but Abd sc fat area was higher in the AA so that the ratio of visceral adipose tissue (VAT) to sc adipose tissue (SAT) areas at L2-3 was lower in AA than in C women, with a similar trend at L4-5 ($P = 0.1$). Glucose at 0 and 120 min after a glucose load, triglycerides, and HDL were similar, but fasting insulin was ∼30% higher in the AA women ($P < 0.01$).

TABLE 1. Subject characteristics

	C		AA		
Baseline	Mean ± SEM (Range)	n	Mean ± SEM (Range)	n	*P*
N	54		31		
Age	59 ± 0.9 (47–77)	54	58 ± 1.3 (49–73)	31	
BMI (kg/m^2)	31.5 ± 0.6 (24–46)	54	34.7 ± 1 (25–45)	31	0.005
Waist-hip ratio	0.82 ± 0.01 (0.67–0.95)	49	0.82 ± 0.01 (0.68–0.97)	27	
$V0_2M$ (ml/min × kg)	20.1 ± 0.6(11–29)	51	17.8 ± 0.95(10–29)	24	0.041
% Fat mass (kg)	46.4 ± 0.8 (30–59)	53	48.6 ± 0.97 (38–58)	30	
Fasting glucose (mg/dl)	94.8 ± 1.1 (79–117)	52	94.9 ± 1.7 (82–122)	31	
Fasting insulin (pM)	73.1 ± 4.3 (27–187)	48	96 ± 8.4 (32–231)	29	0.010
DEXA leg (kg)	15.2 ± 0.6 (6–31)	53	17.8 ± 0.9 (7–29)	31	0.022
DEXA trunk (kg)	19.5 ± 0.7 (11–39)	53	21.8 ± 0.9 (13–33)	31	(0.07)
CT-VAT L2-L3 VAT (cm^2)	162.3 ± 9.2 (79–331)	47	149.8 ± 10.7 (57–296)	28	
CT-VAT L4-L5 VAT (cm^2)	151.9 ± 8.3 (66–310)	48	145.2 ± 10.9 (51–290)	28	
CT-SAT L2-L3 SAT (cm^2)	280.3 ± 16.7 (119–666)	42	360.9 ± 30.6 (192–825)	25	0.014
CT-SAT L4-L5 SAT (cm^2)	403.4 ± 19.8 (124–686)	41	482.8 ± 29.2 (246–851)	25	0.023
CT VAT-SAT L2-L3 ratio	0.61 ± 0.05(0.2–1.7)	39	0.46 ± 0.04 (0.1–0.9)	25	0.039
CT VAT-SAT L4-L5 ratio	0.38 ± 0.03 (0.1–0.7)	39	0.32 ± 0.03 (0.1–0.6)	25	(0.1)
Cholesterol (mg/dl)	197 ± 6 (80–318)	49	200 ± 8 (108–327)	30	
Triglyceride (mg/dl)	116.6 ± 7.3 (37–266)	50	98.5 ± 6.9 (52–172)	30	
HDL (mg/dl)	52.1 ± 1.6(31–78)	50	55 ± 2.3 (32–88)	30	
HOMA-IR	2.9 ± 0.2 (1–7)	47	3.8 ± 0.3 (1–9)	29	0.015

AA women had higher FCW in both depots [race difference: (Glt: 0.60 ± 0.02 (C) 0.68 ± 0.02 (AA) μg lipid/cell; $P < 0.01$); Abd: 0.55 ± 0.02 vs. 0.64 ± 0.02 μg lipid/cell, $P < 0.01$]. FCW was greater in Glt than in Abd [depot difference: $P < 0.001$ (AA), $P < 0.025$ (C)]. There was no difference in the distribution pattern of fat cell sizes between the races (63% of C and 64% of AA displayed a bimodal distribution), and this factor did not affect the lipolysis results (unpublished observation, U.S. and S.K.F.).

Depot differences in lipolysis

Rates of disinhibited lipolysis (ADA alone) did not differ between Abd and Glt adipocytes. PIA, an adenosine receptor agonist, potentially suppressed lipolysis (**Fig. 1**). Rates of lipolysis at 20 nM PIA/cell were lower in Abd than in Glt adipocytes [0.50 ± 0.04 (Abd) vs. 0.58 ± umol/10^6 cells × 2 h (Glt); n = 85; $P < 0.05$; no race interaction). The depot difference was not significant when data were expressed relative to adipocyte surface area to account for the difference in adipocyte size, indicating that the higher lipolysis in Glt adipocytes was simply proportional to the higher adipocyte size. Responsiveness to 20 nM PIA, as determined by calculation of percent suppression, was greater in Abd than Glt sc adipocytes [60% ± 2 (Abd) vs. 54 ± 2% (glt); n = 84; $P < 0.001$], with no interaction with race. Sensitivity to PIA (percent maximal response at 10 or 20 nM PIA) did not differ between the depots (data not shown).

Isoproterenol-stimulated lipolysis was slightly lower in Abd than in Glt adipocytes on a per cell basis [1.7 ± 0.1 (Abd) vs. 2.0 ± 0.1 umol/10^6 cells × 2 h (glt); $P < 0.05$; n = 85, no interaction with race]. The difference was related to the depot difference in fat cell size; expressed per unit adipocyte surface area, rates of maximally stimulated lipolysis were similar in Glt and Abd adipocytes (Fig. 1).

Comparing depots, insulin sensitivity, calculated as ED_{50}, was slightly lower in Glt than in Abd adipocytes of all women combined (19 ± vs. 22 ± 3 pM; n = 58; $P < 0.05$), independent of race. Assessing sensitivity as percent of the maximal response, the depot difference was only apparent at the lowest concentrations of insulin tested: 7.8 pM [37% (Abd) vs. 47% (Glt) in C; $P = 0.05$; and 29 vs. 41% (AA); $P = 0.02$].

Influence of race on lipolysis regulation: higher rates of 'basal' (PIA-suppressed) lipolysis in adipocytes of AA women

On a per cell basis, disinhibited lipolysis was higher in Abd adipocytes of AA than C ($P < 0.03$; independent *t*-test), but this was accounted for by the cell size difference, because it was not apparent when the data were expressed per unit adipocyte surface area (Fig. 1). Rates of basal [PIA-suppressed (20 nM)] per fat cell were ~30% higher in Abd than in Glt adipocytes of AA compared with C ($P < 0.01$), and the difference remained significant in the Abd adipocytes when the data were expressed relative to adipocyte SA ($P < 0.03$). Maximally PIA-suppressed lipolytic rates (100 nM) were higher in Glt adipocytes of AA compared with C, but only when the data were expressed per cell.

To determine whether the race difference in basal lipolysis per unit surface area was due to the hyperinsulinemia or visceral adiposity of the AA, we used two approaches. First, a least square model ($r^2 = 0.12$; $P = 0.02$) showed that fasting insulin ($P < 0.01$) was associated with higher basal lipolysis (log lipolysis at 20 nM PIA) and only a trend toward a race effect ($P = 0.1$). However, in a stepwise multiple regression analysis, the rate of basal lipolysis (log 20 nM PIA value) was associated with both log fasting insulin ($P < 0.01$) and race ($P < 0.05$). Visceral fat, percent body fat, and age did not significantly contribute to either model.

The higher rates of PIA-suppressed lipolysis in Abd adipocytes of AA were associated with a trend toward lower responsiveness [63 ± 3% (C, n = 54) vs. 56 ± 3% suppression

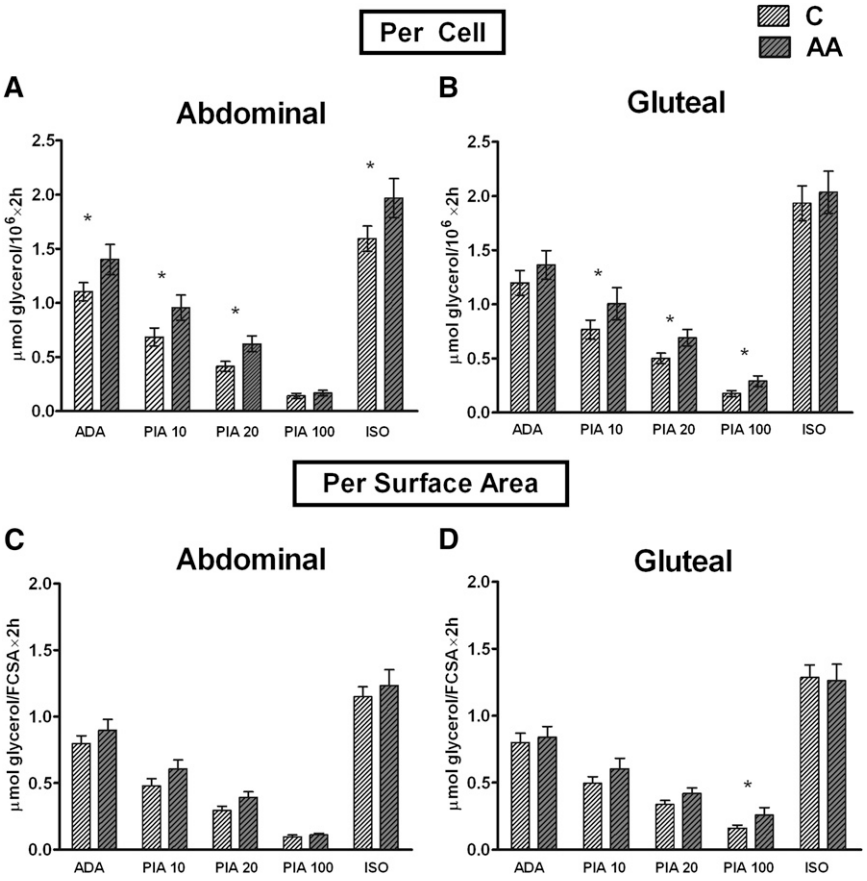

Fig. 1. PIA-suppressed and isoproterenol-stimulated rates in isolated Abd or Glt adipocytes of C (A) or AA (B) women expressed per fat cell cell weight (A,B) or surface area (C,D). Collagenase-isolated adipocytes were incubated with ADA (4 ug/ml) or ADA + 10, 20, or 100 nM PIA, or ADA + 20 nM PIA + 1 µM isoproterenol in Krebs-Ringer bicarbonate buffer containing 4% BSA under an atmosphere of 95% O_2 + 5% CO_2 for 2 h at 37°C. Data are mean ± SEM of 49 C and 29 AA postmenopausal women (except for 10 nM PIA conditions, n = 36 C and 23 AA).

(AA, n = 33; $P = 0.09$)]. Glt adipocytes of AA were significantly less responsive to 20 nM PIA [57 ± 2% (n = 54, C) vs. 49 ± 4% (n = 31, AA); $P < 0.05$].

Stimulated lipolysis did not differ by race

Expressed on a per cell basis, the larger Abd adipocytes of AA compared with C postmenopausal women exhibited ~30% higher rates of stimulated lipolysis in response to a maximal concentration of a nonspecific β adrenergic agonist, isoproterenol ($P < 0.05$). However, there was no race difference in isoproterenol-stimulated lipolysis when the data were expressed relative to adipocyte surface area.

Higher rates of insulin-suppressed lipolysis in Abd adipocytes of AA women

Significant dose-response effects of insulin on lipolysis measured in the presence of ADA were clearly evident in both Abd and Glt adipocytes (insulin effects, $P < 0.001$) (**Fig. 2**). Rates of insulin-suppressed lipolysis were significantly higher in Abd adipocytes of AA women at all submaximal insulin concentrations when the data were expressed on a per fat cell basis (Fig. 2A) and were still evident, but less marked, when the data were expressed relative to adipocyte surface area (Fig. 2C). To determine if the race effect on insulin-suppressed lipolysis was related to the higher body fat or hyperinsulinemia/insulin resistance in the AA group, or related to age, MANOVA analysis (insulin concentration as repeated measure) was performed. The race effect on rates of Abd insulin-suppressed lipolysis per adipocyte (logged values) remained after adjusting for percent fat or regional fat (VAT and SAT area) and age. However, the overall race effect was eliminated in models that included log fasting insulin or homeostasis model of insulin resistance (HOMA-IR). There was a significant interaction of race and insulin concentration, and this was accounted for by the influence of race of the rate of lipolysis at submaximal insulin. Comparing rates of lipolysis at 20 pM insulin, where the magnitude of the race difference is largest (Fig. 2C), there was a trend for a race effect ($P < 0.054$) that was independent of log fasting insulin ($P = 0.04$) in a model that included age and percent fat. Consistent with this finding, responsiveness to 20 pM insulin, calculated as the percent suppression by insulin, was lower in Abd adipocytes of AA than of C women. Regression analysis showed that at 20 pM insulin, percent suppression was affected

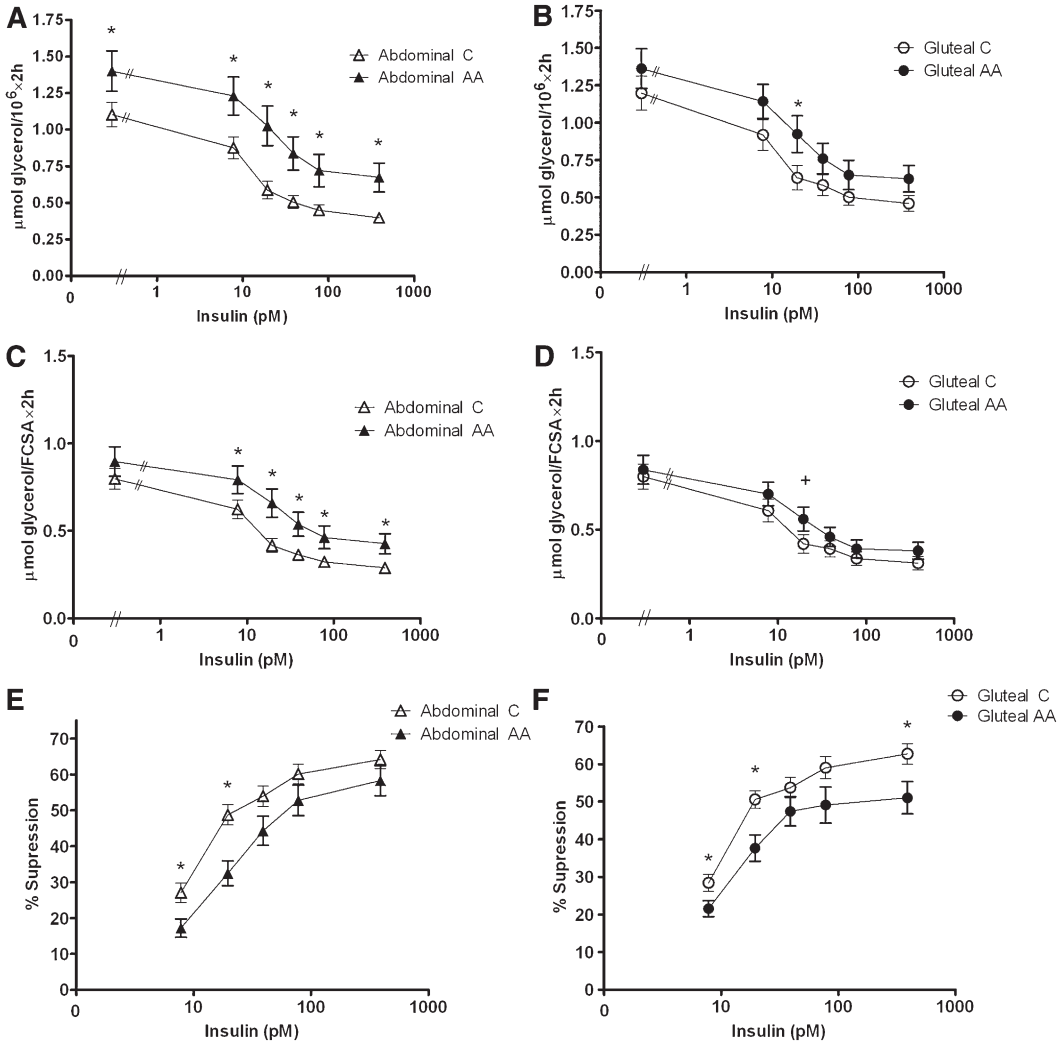

Fig. 2. Decreased insulin-suppressed lipolysis in Abd adipocytes of AA compared with C women. Isolated adipocytes were incubated with ADA or ADA + varying concentrations of insulin (8, 20, 40, 80, or 390 pM) for 2 h, as described in the legend to Fig. 1. Dose response data for Abd or Glt adipocytes from C and AA expressed per million adipocytes (A,B), per unit fat cell surface area (C,D), or percent suppression (E,F). Data are from 49 C and 29 AA postmenopausal women.

by both race ($P < 0.01$) and log fasting insulin ($P < 0.01$) (overall $r^2 = 0.31$; $P < 0.001$). In contrast, at 39 pM insulin, only fasting insulin exerted an significant effect on percent suppression by insulin. These results suggest that the decreased insulin responsiveness of Abd sc adipocytes of AA is mainly associated with the degree of fasting hyperinsulinemia.

To determine whether the decreased response to insulin (percent suppression) at 8 and 20 pM insulin in AA compared with C was related to the trend toward a higher rate of baseline (ADA-stimulated lipolysis), we also calculated the magnitude of the insulin effect as a delta. The delta at 8 and 20 pM insulin was correlated with the baseline (ADA) value. However, at 20 pM insulin, the delta was significantly lower in the AA. Thus, consistent with our conclusion using percent suppression to assess insulin responsiveness, we conclude that AA race is associated with lower responsiveness to submaximal concentrations of insulin.

In contrast to the race effect on insulin responsiveness (percent suppression), there was no statistically significant race difference in insulin sensitivity in Abd adipocytes as assessed by ED_{50} [19.5 ± 3.0 pM (C, n = 38) compared with 27.6 ± 5.0 pM (AA, n = 21) (independent *t*-test of logged values, $P = 0.2$)]. Using percent maximal response as an index of insulin sensitivity, a race effect was apparent only at the lowest insulin concentration tested (8 pM) [27 ± 4% (C, n = 21) vs. 37 ± 3% (AA, n = 35); $P < 0.05$]. This difference did not persist after adjustment for the race difference in log fasting insulin. As expected, log fasting insulin ($r^2 = 0.2$; n = 56; $P < 0.001$) and HOMA-IR ($r^2 = 0.17$; n = 55; $P < 0.002$) were significantly correlated with log ED_{50}, as well as percent maximal response at insulin concentrations of 39 and 78 pM (data from both races combined). Taken together, these results suggest chronic hyperinsulinemia is associated with insulin insensitivity with respect to adipocyte lipolysis (rightward shift in the does response curve) to a similar extent in both races.

We found a significant association between the magnitude of responses to the antilipolytic effect of insulin (percent suppression by 39 pM insulin) in Abd sc adipocytes and either fasting insulin ($r^{2=}$ 0.22; $P < 0.001$; n = 67) or HOMA-IR ($r^2 = 0.26$; $P < 0.001$; n = 65), but no independent effect of race.

No race differences in the insulin-suppressed rates of lipolysis in Glt adipocytes

There was no significant effect of race on lipolysis per surface area or per cell in Glt adipocytes. However, there was a race effect on rates of lipolysis at 19.5 pM insulin by independent Student's *t*-test ($P < 0.05$; logged values; Fig. 2D). Also, consistent with a trend toward a race effect on insulin responsiveness (percent suppression) by MANOVA ($P = 0.07$), posthoc independent *t*-tests showed lower percent suppression in AA at insulin concentrations of 7.8, 19.5, and 390 pM insulin (Fig. 2F). Percent suppression in Glt adipocytes was significantly lower in AA ($P < 0.02$) even when log fasting insulin ($P < 0.001$) was included in a least squares model. Thus, in both depots, there is some evidence for a race effect on responsiveness to insulin at an insulin concentration near the ED_{50}. As in Abd adipocytes, log ED_{50} correlated with log fasting insulin ($r^2 = 0.17$; n = 53; $P < 0.01$) and HOMA-IR ($r^2 = 0.15$; n = 53; $P < 0.005$). There was a trend toward lower insulin sensitivity (log ED_{50}) in Glt adipocytes of AA compared with C ($P = 0.07$).

Race difference in nonesterified FA release from Abd adipocytes

To determine if the higher glycerol release in Abd adipocytes of AA women was paralleled by an increase in FA release, these were measured at selected conditions (0, 7.8, and 390 pM insulin). In Abd adipocytes, FA release per 10^6 fat cells at 390 pM insulin tended to be higher in AA than C (2.42 ± 0.36 vs. 1.57 ± 0.27 μmol/10^6 cell × 2 h; $P = 0.058$), and the race difference was statistically significant when expressed as percent suppression [54.9 ± 4.7% (C) vs. 38.4 ± 2.2% (AA); $P < 0.05$]. The race difference was not statistically significant in a MANOVA analysis when FCW or fasting insulin was in the model. Thus, it appears that the race difference in FA release, similar to glycerol release, is related to both the higher FCW (and surface area) and chronic hyperinsulinemia in the AA women.

DISCUSSION

The current study demonstrated higher rates of insulin-suppressed lipolysis, as measured by glycerol or FA release in Abd sc adipocytes, of overweight postmenopausal AA than C women. These race-related differences persisted when the data were expressed relative to adipocyte surface area to account for the larger adipocytes in AA and were independent of percent fat and visceral adiposity. However, the higher rates of lipolysis in adipocytes of AA were strongly associated with their hyperinsulinemia. Considering relative responsiveness to insulin (percent suppression), AA race was associated with decreased insulin responsiveness, independent of fasting insulin or HOMA-IR. However, there was no race difference in sensitivity to submaximal concentrations of insulin (calculated as ED_{50} or percent maximal response). Our measurement of basal lipolysis (suppressed with 20 nM PIA) was also higher in the AA. This difference was associated hyperinsulinemia but was independent of fat cell size or degree of obesity (percent fat). These in vitro data imply that under conditions that are likely to be physiologically relevant in the postabsorptive state, AA postmenopausal women exhibit higher lipolytic rates. If this alteration at the level of the adipocyte is not compensated for under in vivo conditions, the resulting higher FA flux may contribute to the higher risk for insulin resistance, glucose intolerance, and diabetes in postmenopausal AA compared with C women.

We found that basal lipolysis was ~30% higher on a per cell basis in the larger adipocytes of the AA women. The difference is smaller but statistically significant when expressed relative to adipocyte surface area. Considering FA or glycerol flux across the tissue, the per cell expression is most physiologically relevant. From a mechanistic, more biochemical point of view, however, the race-associated variation in basal lipolysis was minimal. Moreover, multivariate regression analysis indicated that the lower responsiveness to an adenosine agonist was related to the hyperinsulinemia of the AA; that is, it was not attributable to race per se. Nevertheless, the most insulin-resistant women, independent of race, exhibited higher lipolysis under conditions that are likely relevant to lipolysis in vivo, i.e., 10–20 nM PIA is roughly as potent as ~128 nM adenosine, the concentration found in human adipose tissue in vivo by microdialysis (17). This observation is consistent with in vitro studies that show that chronic hyperinsulinemia insulin raises basal lipolysis (18).

In contrast to previous findings in premenopausal women that Glt adipocytes are more sensitive than Abd to antilipolyic effects of insulin in premenopausal women (as assessed by ED_{50}), there was only a very small depot difference in insulin sensitivity in postmenopausal women and was statistically significant only in the AA subjects. Thus, while the enhanced insulin sensitivity of Glt adipocytes to insulin in premenopausal women may favor fat deposition in a lower body pattern, this mechanism may be lost with menopause, most markedly in C women, and therefore contribute to the shift toward a more central deposition of fat that occurs with aging and/or menopause (19).

There were strong negative associations between fasting insulin and sensitivity or responsiveness with the antilipolytic effect of insulin but no race differences in these associations. The downregulation of insulin action secondary to chronic hyperinsulinemia is expected based on in vitro studies (20). This insulin resistance is most apparent at low insulin concentrations, below those found in the hyperinsulinemic subjects, and appears to be completely compensated for by hyperinsulinemia in vivo. For example, Petersen et al. (21) found no difference in systemic or Abd adipose tissue lipolysis (as reflected in glycerol turnover or concentration) under basal or insulin-stimulated conditions when comparing insulin-sensitive and resistant humans based on oral glucose tolerance test results.

The current study provides evidence for a race difference in basal and insulin-suppressed lipolysis in overweight to obese postmenopausal women and contrasts with previous studies of obese premenopausal women in which no race differences, independent of visceral adiposity, were found. Thus, factors associated with age and/or menopause may interact with race-related factors to exacerbate insulin resistance, particularly in Abd sc adipocytes. The decline in sex steroids with menopause may exaggerate a race-related difference in insulin action with secondary effects at the level of adipocyte. Interactions with local paracrine factors or intrinsic differences between Abd and Glt adipocytes may factor into the more marked race-related differences in lipolytic rates and insulin resistance adipocytes of postmenopausal women.

In contrast to the clearly lower rates of insulin-suppressed lipolysis in Abd adipocytes of AA, there were no race-related effects in rate of lipolysis in Glt adipocytes. Nevertheless, expressed as relative response (percent suppression), a lower responsiveness to insulin was also observed in Glt adipocytes of the AA compared with the C women.

Although several previous studies suggested that AA women exhibit low rates of adipocyte lipolysis that may contribute to their propensity toward obesity (11, 22), our data in postmenopausal women, as well as our previously published study of premenopausal women, show that AA race is not associated with a lower rate of lipolysis. In fact, in postmenopausal women, we find the opposite, higher lipolysis under physiologically relevant conditions (i.e., insulin- or PIA-suppressed). Whether a defect in lipolysis exists during the perimenopausal period or during the developmental stages of weight gain during menopause remains to be determined. In this respect, it is noteworthy that lower basal lipolysis (glycerol turnover) was detected in AA compared with C children (23). Nevertheless, our current and previously published data do not support the hypothesis that lower rates of lipolysis contribute to the maintenance of the obese state in pre- or postmenopausal women.

The comparison of group or regional differences in vitro rates of adipocyte lipolysis provides a comparison under highly controlled conditions. The in vitro behavior of adipocytes reflects their prior history (i.e., chronic exposure to hormones such as insulin, local paracrine factors, innervations, and blood flow) as well as genetic influences on lipolytic regulation. The race difference in the insulin suppression of lipolysis in Abd adipocytes was not eliminated by adjusting for insulinemia, but secondary effects of race-related differences in other endocrine or paracrine factors cannot be ruled out.

Our studies only address adipocyte lipolytic capacity and sensitivity to insulin and adenosine. In vivo, however, the balance of endocrine, neural, paracrine, and autocrine factors, as well as innervations and blood flow, will influence the rate of lipolysis. Thus, studies to determine whether the cellular defect in insulin action in adipocytes of AA postmenopausal women has in vivo consequences for systemic FA flux are needed. Albu et al. (8) reported resistance to the antilipolytic effect of insulin in AA premenopausal women, as measured in vivo by the percent suppression of glycerol but not the rate of appearance of FA. However, we found parallel race differences in the insulin suppression of FA and glycerol release from Abd adipocytes of postmenopausal women under the in vitro conditions of our study. We know of no studies that examined potential race differences in in vivo rates of lipolysis and reesterification in postmenopausal women. Studies are needed to assess in vivo FA flux across the abdomen and leg over a range of insulin concentrations in this population. If higher in vivo FA turnover is evident in vivo in AA, the increased availability of FA, combined with a defect in fat oxidation in AA women (24), may contribute to the development of ectopic fat deposition in muscle and liver and thereby the systemic insulin resistance that is observed in postmenopausal AA women (6).

In conclusion, our analyses of in vitro rates of lipolysis in both Abd and Glt fat cells in a relatively large number of healthy, overweight AA and C postmenopausal women show no racial differences in maximal lipolytic capacity (isoproterenol stimulated lipolysis or disinhibited lipolysis). The higher basal lipolysis per adipocyte and decreased relative responsiveness to the antilipolytic effect of insulin in AA than C postmenopausal women may have consequences for metabolism under in vivo conditions. Moreover, the strong association of Abd adipocyte basal lipolysis (submaximally adenosine PIA-suppressed) and insulin insensitivity with circulating insulin, independent of race, suggests that alterations at the level of the adipocyte may contribute to the high systemic FFA flux characteristic of obesity (1).

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