Increased Rates of Lipolysis Among Human Immunodeficiency Virus–Infected Men Receiving Highly Active Antiretroviral Therapy

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Human immunodeficiency virus (HIV) lipodystrophy is associated with fat redistribution, dyslipidemia, and insulin resistance; however, the mechanism of insulin resistance remains unknown. We hypothesized that HIV-infected subjects with fat redistribution have increased rates of lipolysis and increased circulating free fatty acid (FFA) levels that contribute to insulin resistance. Anthrompometric and body composition data were obtained and a standard 75-g oral glucose tolerance test (OGTT) was performed on day 1 of the study. Stable isotope infusions of glycerol and palmitate were completed following an overnight fast to assess rates of lipolysis and FFA flux in HIV-infected men (n = 19) with and without fat redistribution and healthy controls (n = 8) on day 2. Total FFA levels after standard glucose challenge were increased among HIV-infected subjects and positively associated with abdominal visceral adipose tissue area. In contrast, fasting total FFA levels were inversely associated with subcutaneous fat area. Rates of basal lipolysis were significantly increased among HIV-infected subjects (rate of appearance [*Ra*] glycerol, 4.1 ± 0.2 *v* 3.3 ± 0.2 μmol/kg/min in controls; *P* = .02). Among HIV-infected subjects, use of stavudine (*P* = .006) and the rate of lipolysis (ie, *Ra* glycerol, *P* = .02) were strong positive predictors of insulin resistance as measured by insulin response to glucose challenge, controlling for effects of age, body mass index (BMI), waist-to-hip ratio (WHR), and protease inhibitor (PI) exposure. These data demonstrate increased rates of lipolysis and increased total FFA levels in HIV-infected subjects and suggest that increased lipolysis may contribute to insulin resistance in this patient population.

A SYNDROME OF fat redistribution or "lipodystrophy" associated with dyslipidemia and insulin resistance is increasingly recognized among human immunodeficiency virus (HIV)-infected patients and is estimated to affect more than half of all patients receiving combination antiretroviral therapy.[1] The mechanisms responsible for fat redistribution and the associated metabolic disturbances in this syndrome remain uncertain. Accumulating in vitro data suggest a direct effect of protease inhibitors (PIs) interfering with adipogenesis[2] and increasing rates of lipolysis in adipocytes.[2,3] Nucleoside reverse transcriptase inhibitors (NRTIs), in particular stavudine, have also been implicated in the development of fat redistribution[4,5] and have been shown to increase fatty acid β-oxidation in mice.[6]

Higher rates of lipolysis and the resulting increased circulating free fatty acids (FFAs) are one potential mechanism for the development of lipoatrophy and insulin resistance in patients with HIV infection receiving combination antiretroviral therapy. Increased FFA concentrations have been demonstrated among HIV-infected patients with fat redistribution and insulin resistance[7,8]; however, lipolytic rates and the relationship of FFA to insulin resistance have not previously been investigated in this population. To test the hypothesis that increased rates of lipolysis and increased circulating FFAs contribute to insulin resistance in patients with HIV infection and fat redistribution, we evaluated basal lipolytic rates in HIV-infected men and determined the relationship between fat distribution, FFA, and lipolysis. Furthermore, we assessed the potential role of PI and stavudine exposure on lipolysis and insulin resistance in this population.

MATERIALS AND METHODS

Subjects

Nineteen HIV-infected men and 8 healthy men who were similar in age and weight were evaluated in the study between May 2000 and March 2001. Written informed consent was obtained from each subject prior to participation and the study was approved by the Massachusetts General Hospital Human Research Committee and the Massachusetts Institute of Technology (MIT) Committee on the use of Humans as Experimental Subjects.

HIV-infected male subjects between the ages of 18 and 50 years were eligible for the study. Subjects were excluded if they had a previous diagnosis of diabetes; had changed antiretroviral therapy within the past 3 months; were using testosterone, megace, growth hormone, or other anabolic agents within the past 3 months; had a hemoglobin level less than 9.0 g/dL; or had any recent serious opportunistic infections. HIV-infected subjects with and without self-reported changes in fat redistribution were eligible for the study, in order to assess lipolytic rates across a spectrum of body composition changes determined by dual-energy x-ray absorptiometry DEXA and cross-sectional computed tomographic (CT) scanning. Subjects were categorized as having lipodystrophy or no lipodystrophy based on self-report and confirmation by physical examination (single investigator [C.H.]). Lipodystrophy was considered present if a subject had evidence of fat accumulation of the abdomen or neck and/or loss of fat in the face, arms, or legs and had a waist-to-hip ratio (WHR) greater than 0.90. Anitretroviral medication regimen was documented and subjects were later categorized based on current use of PIs and stavudine. Control subjects were healthy men, free of any significant medical conditions, who were matched in age and weight to the HIV infected subjects and had a WHR less than 0.95.

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Pre-infusion Testing

Each subject was instructed to report to the MIT Cancer Research Center (CRC) at 7:30 AM (day 1) following a 12-hour overnight fast (Fig 1) and resting energy expenditure (REE) was determined by indirect calorimetery (Deltatrac, Sensor Medics Corp, Yorba Linda, CA) after a 30-minute rest period. A standard 75-g oral glucose tolerance test (OGTT) was completed with samples obtained at 0, 30, 60, 90, and 120 minutes for insulin and glucose determination. Fasting total, low-density lipoprotein (LDL), and high-density lipoprotein (HDL) cholesterol and triglycerides were measured. HIV-infected subjects also had blood samples collected for CD4 cell count and HIV viral load. All subjects received a standardized caffeine-free diet for the 24 hours prior to the isotope infusion study, which provided 40 kcal/kg body weight with 1 g/kg of protein and 30% to 35% of calories from fat. Subjects were instructed to refrain from strenuous exercise the day prior to the infusion.

Isotope Infusion

Subjects were instructed to complete their evening meal by 9 PM, not to eat or drink thereafter, and to return to the CRC at 8 AM the following morning (day 2). Two intravenous catheters were placed: one for infusion of stable isotopes and one placed retrograde in the hand for blood sampling. The hand was placed in a warming box at 70°C to arterialize the blood. All isotopes were obtained from Masstrace (Woburn, MA). [$^2H_5$] glycerol dissolved in normal saline was infused for 3 hours starting with a priming dose of 1.6 μmol/kg (at time 0) followed by a continuous infusion of 0.11 μmol/kg/min. Similarly, a priming dose of 17.6 μmol/kg of [$^2H_2$] glucose dissolved in normal saline was administered at time 0, followed by a 3-hour continuous infusion of 0.22 μmol/kg/min. [1-^{13}C] palmitate was bound to albumin and administered as a continuous infusion of 0.04 μmol/kg/min over 3 hours. Blood samples for determination of [$^2H_5$] glycerol, [$^2H_2$] glucose, and [1-^{13}C] palmitate enrichment were collected at the following time points after time 0: 45, 60, 75, 90, 120, 150, and 180 minutes. Samples were placed in heparinized tubes and place on ice immediately. Plasma was then separated and stored at -80°C until further analysis.

Body Composition

Cross-sectional abdominal CT scans were performed as described by Borkan et al[9] in order to assess the distribution of subcutaneous and visceral adipose tissue (SAT and VAT, respectively). A lateral scout image was obtained to identify the level of L4 pedicle, which served as the landmark for the 1-cm single-slice image. Subjects also underwent total-body DEXA using the Hologic QDR-4500A scanner (Hologic Inc, Bedford, MA). The DEXA scan was used to determine total body fat and lean body mass. WHR was determined for each subject using measurements of the hip and the waist at the level of the iliac crest taken in triplicate and averaged.

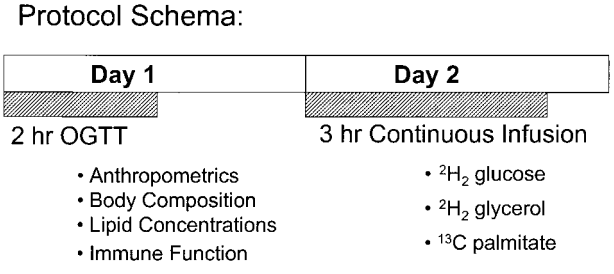

Fig 1. Protocol schema. OGTT, 75-g oral glucose tolerance test. For details of infusion protocol and priming doses, see text.

Calculations

The rate of appearance (*Ra*) of glycerol or palmitate in plasma was calculated using Steele's equation[10] as it applies to isotopic steady-state conditions. Therefore, *Ra* (μmol/kg/min) = F/IE, where F is the isotopic infusion rate (in μmol/kg/min) and IE is the isotopic enrichment at plateau. Due to the infusion of these isotopes and the relatively smaller pool size, contributing approximately 2% of the substrate pool, the above equation can be modified to: *Ra* (μmol/kg/min) = (IE_i /IEp – 1) · F, where *Ra* is the rate of appearance of glycerol or palmitate, IEi is the isotopic enrichment of the infusate (atom percent excess [APE]), and IEp is the isotopic enrichment in plasma (APE) at isotopic equilibrium.

At isotopic equilibrium, assuming substrate concentrations are stable, Substrate uptake = *Ra* (μmol/kg/min) and Substrate (palmitate or glycerol) clearance rate = Substrate uptake/Substrate plasma concentration.

Since palmitate is thought to be typical of other long-chain fatty acids, the flux of all fatty acids is assumed to be similar to that of palmitate.[11]

Homeostatic model to assess insulin resistance (HOMA-IR)[12] was calculated from the determination of the fasting insulin and glucose levels obtained on the day of the infusion. Insulin and glucose areas under the curve (AUCs) were calculated from the values obtained at 0, 30, 60, 90, and 120 minutes following oral glucose challenge performed on the day prior to the assessment of lipolytic rates.

Biochemical and Immunologic Assays

Levels of glucose, cholesterol, triglyceride, HDL, and LDL were determined by methods previously reported.[13] Nonesterified or "free" fatty acid (FFA) concentrations were measured using an in vitro enzymatic colorimetric assay kit (Wako Chemicals USA, Inc, Richmond, VA). The intra-assay coefficient of variation for FFA ranged from 1.1% to 2.7%. The published normal range for FFA is 0.1 to 0.6 mmol/L. Insulin levels were measured in serum by radioimmunoassay (Diagnostic Product Corp, Los Angeles, CA). Intra-assay and interassay coefficients of variation range from 4.7% to 7.7% and 5.5% to 9.2%, respectively. CD4 count was determined by flow cytometry (Becton Dickinson Immunocytochemistrty Systems, San Jose, CA) and HIV viral load was determined by ultrasensitive assay (Amplicor HIV-1 Monitor Assay, Roche Molecular Systems, Branchburg, NJ) with limits of detection 50 to 75,000 copies/mL.

Statistical Analyses

Univariate clinical characteristics are presented as means ± SEM. Between-group comparisons were made using Student's *t* test. Analysis of variance (ANOVA) was used to evaluate potential group differences in insulin AUC. Data from one control subject were dropped from the calculations of *Ra* of glycerol due to unexplained variability in glycerol enrichment calculations. Multivariate regression analyses were completed to evaluate predictors of lipolysis and insulin resistance, and all models included age, body mass index (BMI), WHR, lipodystrophy (coded yes or no), stavudine exposure (coded yes or no), and PI exposure (coded yes or no). Subgroup comparisons were made among HIV-infected subjects based on lipodystrophy, PI, and stavudine status using Student's *t* test. To correct for multiple subgroup comparisons, a *P* value of less than .01 was used to determine statistical significance. All statistical analyses were performed using SAS JMP (SAS Institute, Inc, Cary, NC) and statistical significance was defined as a 2-tailed alpha value of *P* less than .05 unless otherwise stated.

RESULTS

Nineteen HIV-infected men and 8 healthy volunteers matched for age and body mass index completed the protocol.

The demographic characteristics, body composition and energy expenditure parameters of the subjects are summarized in Table 1. HIV-infected subjects had higher mean REE, triglyceride concentration, HOMA-IR, glucose AUC following OGTT and FFA AUC following OGTT compared to control subjects (Table 2). In addition, HIV-infected subjects had significantly lower HDL cholesterol compared to controls. Although not statistically significant, there were trends towards lower mean percent body fat (19 ± 1 *v* 23 ± 2%, $P = 0.05$), higher WHR (0.95 ± 0.02 vs 0.90 ± 0.02, $P = 0.09$) and higher insulin AUC after OGTT (7,342 ± 987 vs 3,969 ± 528 μU/mL×120 min, $P = 0.06$) among the HIV-infected men compared to controls. The lower mean percent body fat in HIV-infected subjects was attributable to lower, but not statistically significant, subcutaneous adipose tissue compared to controls (HIV *v* control: 12,611 ± 1,379 mm^2 *v* 17,820 ± 3,277 mm^2, $P = 0.09$).

Among all subjects, FFA AUC was positively correlated with insulin AUC following OGTT (r=0.45, $P = 0.02$). This association was also significant among HIV-infected subjects only (r=0.51, $P = 0.04$). There was no significant effect of group (HIV with lipodystrophy, HIV without lipodystrophy and control) on insulin AUC ($P = 0.07$).

HIV-infected subjects had significantly increased basal rates of lipolysis compared to control subjects (HIV vs control: *Ra* of glycerol: 4.1 ± 0.2 *v* 3.3 ± 0.2 μmol/kg/min, $P = 0.02$, see Table 2). *Ra* glycerol per kg of body fat was also significantly higher in the HIV-infected subjects (HIV vs control: 23.0 ± 1.8 *v* 15.6 ± 2.4 μmol/kg body fat/min, $P = 0.03$). There was no difference between HIV-infected subjects and controls in the *Ra* of glucose. FFA flux as estimated from *Ra* palmitate was not statistically different between subject groups.

Visceral adipose tissue area (VAT) of the abdomen was positively associated with increased post-glucose challenge FFA concentrations (all subjects r=0.51, $P = 0.006$ and among HIV subjects only r=0.58, $P = 0.01$). In contrast, subcutaneous adipose tissue area (SAT) was inversely correlated with fasting FFA levels (all subjects r= -0.48, $P = 0.01$ and HIV subjects only r= -0.51, $P = 0.02$). There was no significant correlation detected between measures of regional body fat by DEXA, VAT, SAT or VAT:TAT ratio by CT scan and *Ra*'s for glycerol, glucose or palmitate or insulin AUC.

Table 1. Characteristics of HIV-Infected Subjects and Healthy Control Subjects: Body Composition and Energy Expenditure

	HIV (n = 19)	Control (n = 8)
Age (yr)	40 (1)	40 (3)
Duration of HIV (yr)	8.5 (1.0)	—
HIV viral RNA (copies/mL)	14,035 (5,937)	—
CD4 count (cells/μL)	418 (62)	—
Height (cm)	175.4 (1.2)	172.7 (3.0)
Weight (kg)	75.7 (1.8)	74.8 (4.1)
BMI (kg/m^2)	24.6 (0.5)	25.0 (1.0)
WHR	0.95 (0.02)	0.90 (0.02)
Fat mass (kg)	14.3 (1.0)	17.7 (2.1)
Fat free mass (kg)	61.8 (1.5)	57.8 (2.9)
% Body fat	19 (1)	23 (2)
VAT (mm^2)	8,896 (1,521)	6,659 (1,220)
SAT (mm^2)	12,611 (1,379)	17,820 (3,277)
VAT:TAT	0.38 (0.05)	0.30 (0.04)
Resting energy expenditure (kcal/d)	1,854 (64)*	1,544 (59)
Respiratory quotient	0.82 (0.01)	0.83 (0.02)

NOTE. Values are mean (SEM); fat mass, fat free mass, and % body fat were obtained from total-body DEXA.

*$P < .01$.

Table 2. Characteristics of HIV-Infected Subjects and Healthy Control Subjects: OGTT, Lipid and Basal Lipolytic Rates

	HIV (n = 19)	Control (n = 8)
HOMA-IR	6.5 (0.5)†	4.9 (0.3)
Fasting FFA (mmol/L)	0.50 (0.04)	0.38 (0.06)
Insulin AUC (μU/mL × 120 min)	7,342 (987)	3,969 (528)
Glucose AUC (mg/dL × 120 min)	17,353 (968)*	12,148 (816)
FFA AUC (mmol/L × 120 min)	35.5 (3.2)†	24.4 (2.4)
Cholesterol (mg/dL)	191 (12)	169 (19)
Triglycerides (mg/dL)	188 (28)†	70 (9)
LDL (mg/dL)	116 (10)	103 (18)
HDL (mg/dL)	40 (3)†	53 (5)
Ra glycerol (μmol/kg/min)	4.1 (0.2)†	3.3 (0.2)
Ra palmitate (μmol/kg/min)	1.3 (0.1)	1.2 (0.2)
Ra glucose (μmol/kg/min)	13.0 (0.5)	13.4 (.6)

NOTE. Values are mean (SEM).

*$P < .01$ and †$P < .05$, *t* test results: HIV *v* control.

HIV-Infected Subgroup Comparisons

Subanalyses were performed on HIV-infected subjects based on the presence of lipodystrophy, and current exposure to a protease inhibitor (PI) and stavudine (Table 3). Eleven subjects were currently on a PI containing regimen and 11 subjects were currently on stavudine; 5 subjects were on both. Compared to subjects without lipodystrophy, subjects with lipodystrophy had significantly increased VAT, VAT to total adipose tissue (TAT) ratio, and increased trunk percent fat by DEXA. Subjects on a PI-containing regimen had increased SAT and increased trunk percent fat by DEXA. There were no differences in insulin or glucose measurements or the *Ra*'s for glycerol, glucose or palmitate based on lipodystrophy or PI status. However, subjects currently on stavudine had significantly increased insulin AUC after glucose challenge, as well as increased fasting *Ra* of palmitate (ie, FFA flux) compared to subjects not currently on stavudine. Furthermore, subjects on stavudine had significantly increased VAT and VAT:TAT ratio compared to those not on stavudine, indicative of substantial fat redistribution.

Predictors of Lipolysis and Insulin Resistance

Multivariate regression analyses were used to evaluate predictors of lipolysis and insulin resistance (assessed by insulin AUC and HOMA-IR) among HIV-infected subjects. In a model including age, BMI, WHR, presence or absence of lipodystrophy, PI use, and stavudine use, there were no statistically significant predictors of fasting rates of lipolysis (*Ra* glycerol). Use of stavudine ($P = .03$), however, was predictive of increased FFA flux (*Ra* palmitate) (whole-model $r^2 = .54$, stavudine use estimate = 0.4; 95% confidence interval [CI], 0.06 to

Table 3. Metabolic Parameters Among HIV-Infected Subjects According to Lipodystrophy Status, PI Use, and Stavudine Use

	Lipodystrophy (n = 11)	No Lipodystrophy (n = 8)	Current PI Use (n = 11)	No Current PI Use (n = 8)	Current Stavudine Use (n = 8)	No Current Stavudine Use (n = 11)
Age (yr)	42 (2)	37 (2)	41 (1)	38 (2)	43 (2)	38 (2)
BMI (kg/m^2)	24.8 (0.8)	24.3 (0.7)	25.1 (0.6)	23.8 (0.8)	24.7 (0.8)	24.5 (0.7)
Duration of HIV (yr)	9.4 (1.0)	7.3 (2.0)	7.5 (1.3)	9.9 (1.5)	9.1 (1.2)	8.1 (1.5)
Current PI use (%)	64%	50%	100%	0%	62%	55%
Current stavudine use (%)	64%	12%	45%	38%	100%	0%
CD4 count (cells/μL)	461 (64)	360 (120)	441 (71)	388 (114)	444 (71)	400 (96)
VAT (mm^2)	12,791 (1,862)*	3,541 (490)	8,274 (1,623)	9,752 (2,963)	13,459 (2,526)*	5,578 (1,154)
SAT (mm^2)	13,243 (2,117)	11,742 (1,621)	15,803 (1,584)*	8,221 (1,389)	11,203 (2,065)	13,635 (1,869)
VAT:TAT	0.49 (0.06)*	0.23 (0.02)	0.32 (0.04)	0.46 (0.1)	0.53 (0.09)*	0.28 (0.02)
WHR	0.99 (0.02)*	0.89 (0.02)	0.96 (0.02)	0.92 (0.03)	1.0 (0.02)*	0.90 (0.02)
% Body fat	19 (2)	18 (1)	20 (1)	16 (1)	18 (1)	19 (2)
Trunk % fat	22 (2)*	16 (2)	22 (2)†	16 (2)	20 (2)	19 (2)
Extremity % fat	15 (2)	19 (1)	18 (2)	15 (2)	15 (2)	18 (2)
HOMA-IR	6.9 (0.7)	6.0 (0.7)	7.1 (0.6)	5.8 (0.8)	7.9 (0.9)†	5.6 (0.3)
Fasting FFA (mmol/L)	0.51 (0.06)	0.47 (0.07)	0.47 (0.04)	0.50 (0.1)	0.56 (0.07)	0.45 (0.06)
Insulin AUC (μU/mL $\times$ 120 min)	8,292 (1,358)	6,036 (1,381)	7,811 (1,330)	6,698 (1,541)	10,299 (1,762)*	5,192 (593)
Glucose AUC (mg/dL $\times$ 120 min)	18,522 (1,027)	15,516 (1,772)	16,845 (1,132)	17,989 (1,719)	19,056 (990)	15,992 (1,453)
FFA AUC (mmol/L$\times$120 min)	41.3 (4.4)	27.6 (3.2)	33.0 (2.3)	39.0 (7.1)	42.9 (6.2)	30.1 (2.5)
Fasting *Ra* glycerol (μmol/kg/min)	4.0 (0.2)	4.1 (0.3)	4.2 (0.2)	3.9 (0.3)	4.0 (0.3)	4.2 (0.2)
Fasting *Ra* glucose (μmol/kg/min)	13.1 (0.6)	13.0 (0.8)	12.9 (0.6)	13.2 (0.8)	12.7 (0.8)	13.3 (0.6)
Fasting *Ra* palmitate (μmol/kg/min)	1.4 (0.08)	1.2 (0.1)	1.4 (0.8)	1.2 (0.1)	1.5 (0.08)*	1.1 (0.07)

NOTE. Values are mean (SEM).

*$P < .01$ and †$P < .05$, *t* test results of comparison between lipodystrophy *v* no lipodystrophy, PI use *v* no PI use, and stavudine use *v* no stavudine use.

0.77). Using a similar model, *Ra* glycerol ($P = .02$) and use of stavudine ($P = .006$) were strong positive predictors of HOMA-IR, independent of the effects of age, BMI, WHR, lipodystrophy, and PI use (whole-model $r^2 = .69$, *Ra* glycerol estimate = 1.4 [95% CI, 0.2 to 2.6]; stavudine use estimate=3.5 [95% CI, 1.2 to 5.8]). Similarly, *Ra* glycerol ($P = .03$) and stavudine use ($P = .003$) were independent positive predictors of insulin AUC following glucose challenge (whole-model $r^2 = .67$, *Ra* glycerol estimate = 2,882 [95% CI, 356 to 5,407]; stavudine use estimate = 8,330 [95% CI, 3,599 to 13,061]). *Ra* palmitate, when added to the model, was not a significant predictor of HOMA-IR or insulin AUC, but *Ra* glycerol and stavudine use remained significant.

DISCUSSION

Metabolic abnormalities, including insulin resistance, dyslipidemia, and changes in fat distribution are a significant problem for many HIV-infected patients receiving highly active antiretroviral therapy. Although specific medications may affect glucose homeostasis directly,[14] it is equally plausible that substantial changes in fat distribution or direct effects of certain medications on rates of lipolysis contribute to insulin resistance in these patients. Increased lipolysis, either as a result of excess visceral adiposity, or as the mechanism of peripheral fat loss via the influence of antiretroviral medications, may lead to elevations in circulating FFA concentrations. Previous investigation demonstrated that FFA levels are increased in HIV-infected patients with fat redistribution.[7,8] The current study investigated lipolysis, FFA flux, and the link between increased FFA and insulin resistance in HIV-infected patients. Our data demonstrate that abnormal lipolysis and increased FFA, associated with abnormal fat distribution and use of stavudine, contribute significantly to insulin resistance in HIV-infected patients.

In a preliminary report, Morlese and et al[15] used isotope techniques to evaluate lipid metabolism in HIV-infected subjects before and after the initiation of PI therapy. The rate of lipid oxidation, measured following an oral palmitate load, increased in patients after 3 months of PI therapy compared to baseline. These data, combined with in vitro observations of increased lipolysis in adipocytes exposed to PIs,[2,3] suggest that HIV protease inhibitors may directly contribute to the development of clinical lipodystrophy as well as its associated metabolic disturbances. In the current study, however, while PI use was associated with increased SAT and rising cholesterol levels, PI use was not associated with increased rates of lipolysis, FFA flux, or circulating FFA concentrations. Furthermore, PI use was not associated with increased indices of insulin resistance in this population.

The use of stavudine, a NRTI, has been shown to result in decreased subcutaneous fat and other metabolic disturbance in HIV-infected patients on highly active antiretroviral therapy.[4] Furthermore, stavudine exposure resulted in increased hepatic fatty acid oxidation in lean mice and decreased white adipose tissue mitochondrial DNA levels in obese mice.[6] Our study demonstrated that HIV-infected patients currently on stavudine had significantly more VAT and higher VAT:TAT ratios compared to patients not on stavudine, and these same patients had increased insulin response to glucose challenge. Furthermore, we show that stavudine use was a strong independent predictor of insulin AUC and HOMA–IR controlling for age, BMI, WHR, and PI status. Use of stavudine was associated with a

113% increase in insulin AUC among HIV-infected subjects. In addition, stavudine users had increased fasting levels of FFA flux compared to those not taking stavudine. While these findings do not establish causality, our data agree with previous clinical and animal observations[4-6] and suggest that stavudine may have a direct effect on peripheral and visceral fat metabolism, which in turn influences FFA levels and resultant insulin resistance.

FFA flux as measured by palmitate turnover was not statistically significantly elevated in HIV-infected subjects, despite finding significant increases in total circulating FFA concentrations and increased rates of lipolysis compared to control subjects. Palmitate is used as a representative marker for overall fatty acid metabolism, but in this study of relatively small sample size, palmitate may not have been a sensitive enough surrogate to detect differences in total FFA metabolism between subject groups.

One possible explanation for current findings is that HIV infection may have a direct influence on lipid metabolism and accounts for the increased rates of lipolysis observed in the HIV-infected group compared to controls. Prior to the recognition of lipodystrophy syndrome, increased triglyceride and FFA concentrations as well as decreased triglyceride clearance were identified among HIV-infected patients with acquired immunodeficiency syndrome (AIDS).[16] Viral influences may directly alter adipocyte metabolism and/or hepatic regulation of lipid metabolism. Other potential mechanisms for increased lipolysis among HIV-infected patients may include changes in cytokines such as tumor necrosis factor-alpha (TNFα), and adipocyte regulatory hormones such as ACRP30 and resistin, which may be influenced by antiretroviral medications, changes in fat distribution, or HIV itself. Furthermore, it is possible that subjects without overt evidence of fat redistribution may experience alterations in metabolism and lipolysis related to medication use or HIV that is either independent of or precedes clinically apparent fat redistribution.

In summary, we demonstrate increased rates of lipolysis among HIV-infected subjects and show that increased lipolysis is associated with insulin resistance in this population. Further, circulating FFA concentrations, which may interfere with insulin and glucose homeostasis, are increased in association with fat redistribution among HIV-infected patients. Future investigation is necessary to further elucidate the mechanistic links between lipolysis, fatty acid metabolism, insulin resistance, and fat redistribution in patients with HIV infection and lipodystrophy.

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