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Antiretroviral Drug Levels and Interactions Affect Lipid, Lipoprotein, and Glucose Metabolism in HIV-1 Seronegative Subjects: A Pharmacokinetic-Pharmacodynamic Analysis

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ABSTRACT

Background: **HIV-infected patients treated with antiretroviral medications (ARVs) develop undesirable changes in lipid and glucose metabolism that mimic the metabolic syndrome and may be proatherogenic. Antiretroviral drug levels and their interactions may contribute to these metabolic alterations.**

Methods: **Fifty six HIV-seronegative adults were enrolled in an open-label, randomized, pharmacokinetic interaction study, and received a nonnucleoside reverse transcriptase inhibitor (efavirenz on days 1–21) plus a protease inhibitor (PI; amprenavir on days 11–21), with a second PI on days 15-21 (saquinavir, nelfinavir, indinavir, or ritonavir). Fasting triglycerides, total LDL- and HDL-cholesterol, glucose, insulin, and C-peptide levels were measured on days 0, 14, 21, and 2–3 weeks after discontinuing drugs. Regression models were used to estimate changes in these parameters and associations between these changes and circulating levels of study drugs.**

Results: **Short-term efavirenz and amprenavir administration significantly increased cholesterol, triglycerides, and glucose levels. Addition of a second protease inhibitor further increased triglycerides, total and LDL-cholesterol levels. Higher amprenavir levels predicted larger increases in triglycerides, total, and LDL-cholesterol. Two weeks after all study drugs were stopped, total, LDL-, and HDL-cholesterol remained elevated above baseline.**

Conclusions: **ARV regimens that include a nonnucleoside reverse transcriptase inhibitor plus single or boosted PIs are becoming more common, but the pharmacodynamic interactions associated with these regimens can result in persistent, undesirable alterations in serum lipid/lipoprotein levels. Additional pharmacodynamic studies are needed to examine the metabolic effects of ritonavir-boosted regimens, with and without efavirenz.**

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INTRODUCTION

Forty to 60% of HIV-infected patients treated with antiretroviral medications (ARVs) experience undesirable metabolic, endocrine, and body composition changes, including insulin resistance, dyslipidemia, elevated diastolic blood pressure, visceral adiposity and/or peripheral lipoatrophy, elevated serum biomarkers for prothrombotic events, and chronic inflammation.[1–10] These are components of the cardiometabolic syndrome, and threaten to increase cardiovascular events in HIV-infected people,[3,4,11–26] but little is known about how ARV drug levels and interactions contribute to these metabolic alterations.

Many factors contribute to metabolic alterations in HIV-infected people treated with ARVs: HIV-1 infection, direct and indirect actions of ARVs on substrate metabolism, genetics, cytokines, nutrition, physical inactivity, behavior, gender, and age. Most studies have focused on HIV-protease inhibitors (PIs) as causative factors. In healthy HIV-seronegative subjects, short-term exposure (≤4 weeks) to the PI indinavir impaired peripheral and hepatic insulin sensitivity without affecting lipid/lipoproteins,[27,28] whereas short-term lopinavir/ritonavir exposure impaired insulin sensitivity,[29] and increased serum triglycerides (TG), very low-density lipoprotein-cholesterol (VLDL-c), and free fatty acid levels without changing low- or high-density lipoprotein-cholesterol (LDL-c or HDL-c).[30] Two-week exposure to escalating doses of ritonavir increased TG and VLDL-c (but not LDL-c), and reduced HDL-c in HIV-negative subjects.[31] Five days of atazanavir exposure, however, did not affect insulin sensitivity.[29] In general, indinavir-containing regimens tend to alter glucose metabolism, while ritonavir-containing regimens tend to prompt dyslipidemia. These studies are limited because individual PIs were administered. In current clinical practice, regimens include dual- or boosted-PIs plus other ARV drug classes (nucleoside and nonnucleoside reverse transcriptase inhibitors, NRTI, NNRTI). Little is known about the direct pharmacokinetic interactions among these drug combinations and their effects on serum lipid and glucose parameters.

Large HIV-infected cohorts corroborate these findings.[32,33] PI-containing regimens (especially ritonavir) were associated with higher TG and total cholesterol (TC) levels than indinavir-containing regimens, or non-PI-containing regimens. NNRTI-containing regimens (efavirenz, nevirapine) were associated with higher TC and LDL-c values than those in ARV-naive patients. Efavirenz- and nevirapine-containing regimens have been associated with increments in HDL-c levels,[33–36] despite their undesirable effects on serum TG and TC levels. Nevirapine-containing ARV regimens have been associated with larger increases in HDL-c and decreases in the ratio of TC–HDL-c (TCHC) than efavirenz-containing regimens.[37] Taken together, these findings suggest that some PI-to-NNRTI substitutions and PI-sparing regimens may be associated with more favorable lipid profiles in HIV-infected people.

We hypothesized that alterations in fasting lipid/lipoprotein and glucose metabolism are caused by combined actions of efavirenz (NNRTI) and amprenavir (PI), compounded by the addition of a second PI through pharmacokinetic (PK) drug interactions, and that markers of lipid and glucose metabolism would be related to plasma efavirenz and amprenavir concentrations. To test these hypotheses, fasting serum lipid/lipoprotein, glucose, insulin, and C-peptide levels and drug concentrations were quantified during a pharmacokinetic interaction study conducted in healthy, HIV-1-seronegative men and women exposed to a short course of efavirenz plus amprenavir, followed by the addition of a second PI (saquinavir, nelfinavir, indinavir, ritonavir, or none/control).

METHODS

Study design

ACTG A5043 was an open-label, randomized study. Subjects received a single, oral dose of amprenavir 600 mg alone on day 0, efavirenz 600 mg daily alone on days 1-10, amprenavir 600 mg once every 12 hours (q12h) plus efavirenz 600 mg q24h on days 11-14, and efavirenz plus amprenavir with or without administration of a second, randomly assigned PI on days 15–21 (Figure 1). The second PI was

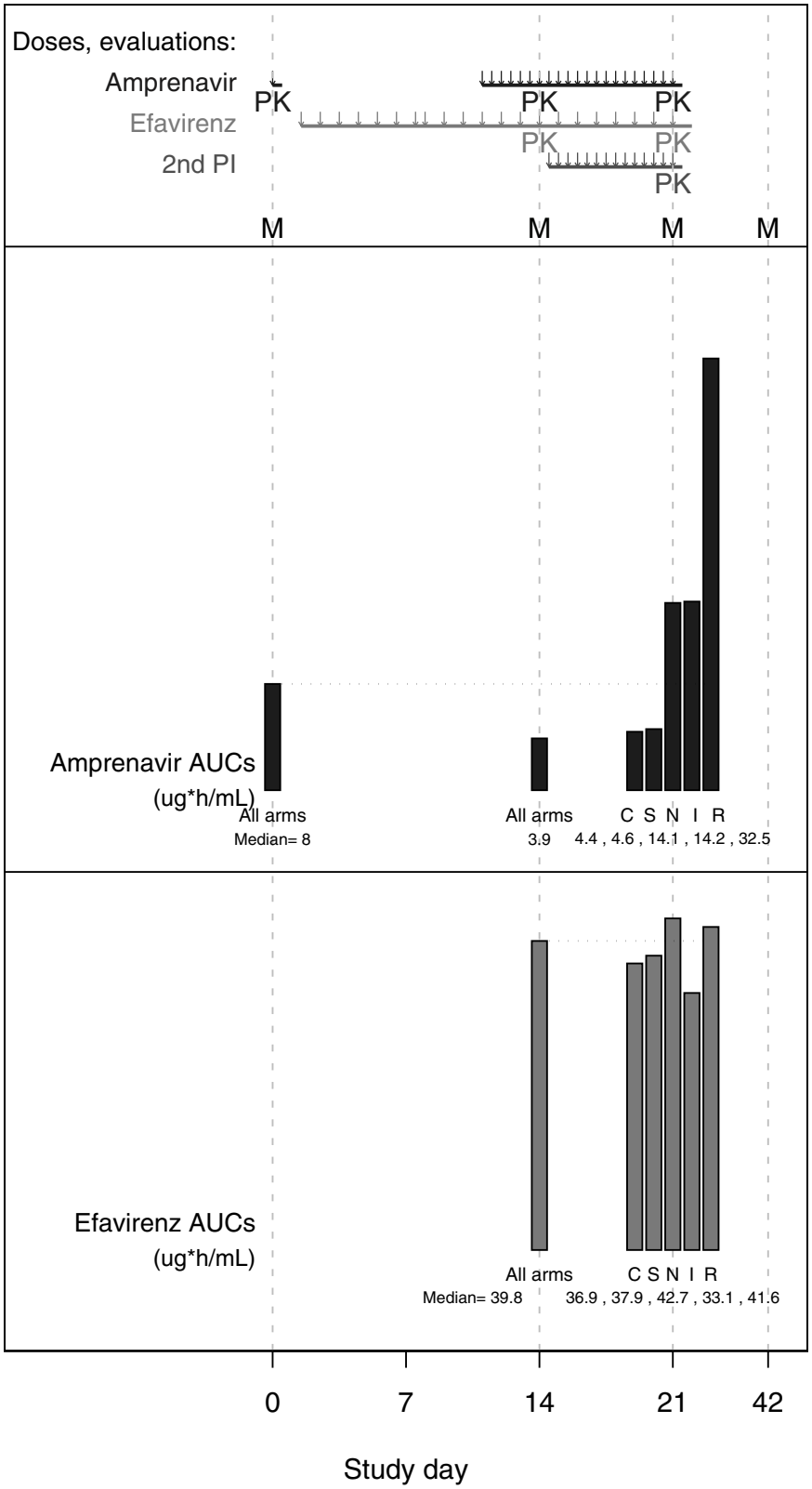

FIG. 1. Top panel: Schedule of ACTG A5043 study drug administration and laboratory evaluations (PK, intensive pharmacokinetic sampling conducted; M, fasting metabolic samples collected). Middle panel: Summary of amprenavir PK findings. Top of bar and number indicate median AUC. On days 0 and 14, all arms are combined. On day 21, arms are presented separately: C, control; S, saquinavir; N, nelfinavir; I, indinavir; and R, ritonavir. Bottom panel: Summary of efavirenz PK findings.

saquinavir soft gelatin capsules 1600 mg q12h, nelfinavir 1250 mg q12h, indinavir 1200 mg q12h, ritonavir 100 mg q12h, or none (control arm). One objective was to compare the PK of amprenavir when taken alone, with efavirenz, and with efavirenz plus a second PI; and the PK of efavirenz taken with amprenavir, with and without a second PI. A second objective was to examine the effects of these drug combinations/interactions on fasting glucose and lipid metabolism.

Inclusion criteria included HIV-1-seronegative, 18–65 yr old, male or female, ≥ 50 kg and within 20% of ideal body weight, of stable health over the preceding 6 months, liver function tests $<1.25 \times$ upper limit of normal (ULN), and fasting TC and TG ≤ 200 mg/dL. Other study details are described elsewhere.[38] The purpose, risks, benefits, and confidentiality concerns were explained to each volunteer. All eligible volunteers signed an IRB-approved informed consent document.

On days 0 (baseline, prior to exposure to any study drug), 14, and 21, and 2–3 weeks after discontinuation of all study drugs (day 42), fasting blood samples (7.5–16.5h from food/drink intake) were collected in the morning, prior to administering study drug. Samples were stored at −20°C and analyzed for TG, TC, HDL-c, glucose (GLU), insulin (INS), and C-peptide (CPEP) levels in batches when the study ended in site-designated laboratories. Subject height and weight were measured on day 0; weight was also measured on days 14, 21, and 42. The following measures were calculated: LDL-c (TC-[HDL-c + {TG = 5}]), the ratio of TC to HDL-c (TCHC), the homeostasis model assessment of insulin resistance (HOMA = fasting GLU [mM] × fasting INS [μU/mL]/22.5),[39] and body mass index (BMI = weight [kg]/height [m^2]). For 5 of 222 subject-day occasions (2%), GLU, INS, CPEP, and TG values were clearly nonfasting, and were excluded from analysis. PK sampling was conducted on days 0, 14 and 21. Compliance was assessed by pill counts and self-report. Subjects were excluded from analyses if ≥1, ≥2 or ≥6 doses were missed in the 48h, 72h or 14 days, respectively, prior to PK/metabolic specimen collection. For a given lipid/glucose measure, a subject's data were included if the subject contributed valid values on ≥2 occasions. NCEP ATP III guidelines for the metabolic syndrome[40] were used to classify fasting TC, LDL-c, HDL-c, and TG values. Fasting GLU >110mg/dL and INS levels >15μU/mL were considered elevated.

Statistical analysis

Baseline values. Descriptive statistics are reported by study day. ANOVA models were used to evaluate baseline differences in metabolic parameters, weight, height, and BMI by treatment arm and race/ethnicity (African-American/nonHispanic vs others).

Changes over time. We focused on three comparisons: (1) day 14 vs. baseline, i.e., how metabolic parameters changed after 14 days of efavirenz and 3 days of amprenavir exposure; (2) day 21 vs. 14, i.e., how parameters changed after 7 additional days of efavirenz and amprenavir plus (in four arms) exposure to a second PI; and (3) day 42 vs. baseline, i.e., metabolic parameters 2–3 weeks after discontinuing all medications. A repeated-measures model was fit to available data for a given measure, consisting of an overall mean (day 0 values); additive fixed effects for days 14, 21 and 42; and a random intercept for each subject.

For a given outcome, if day was statistically significant, least-squares means of changes over time were reported, and considered statistically significant if associated 95% CIs excluded the value zero. For outcomes where day was significant, additional differences between PIs (arms) on day 21 were evaluated. Where day was not significant, indicators for presence/absence of each second PI were evaluated as the only model covariates.

Relationships between ARV PK parameters and metabolic alterations. Compared to amprenavir areas under the curve (AUCs) when taken alone (day 0), AUCs of amprenavir taken with efavirenz (day 14) were smaller, and taken with efavirenz and a second PI were more variable[38] (Figure 1). On day 21, amprenavir AUCs were highest for subjects taking ritonavir and lowest on the control and saquinavir arms.[38] Regression models were used to estimate effects of

day 14 amprenavir AUC and baseline metabolic value on metabolic changes at day 14. If amprenavir AUC was a significant explanatory variable, the regression equation was reported, along with predicted metabolic outcomes over a range of possible amprenavir exposures. Effects of day 21 amprenavir AUCs on day 21 metabolic changes, and of efavirenz AUCs on both days, were similarly assessed.

Deviance statistic p-values[41] ≤ 0.05 were considered significant. Analyses were exploratory; therefore, no adjustments were made for multiple comparisons.

RESULTS

Of 82 subjects enrolled into the study and receiving some study drug, 56 took drug per protocol, had valid results for at least one metabolic outcome, and were therefore included in analyses. Numbers of subjects included in the analyses relative to the number randomized to each second PI arm were as follows: control 11/16, saquinavir 10/16, nelfinavir 12/18, indinavir 13/19, and ritonavir 10/16. Baseline weight and BMI did not differ significantly by arm. Body weight fluctuations over the 42 days were minor, ranging from −6% to +8% (±5 kg). There were no differences in body weight or BMI by study day. Baseline metabolic outcomes did not differ by race/ethnicity (African-American/non-Hispanic vs. other).

Baseline lipid, lipoprotein, and glucose parameters are given in Table 1. TC and LDL-c were within the normal range at baseline for all subjects. At baseline, 18% of subjects had TG above the NCEP threshold (150 mg/dL); 33% of 49 men and 0% of the 2 women had HDL-c below their respective thresholds of 40 and 50 mg/dL. Baseline CPEP was lowest in subjects randomized to ritonavir and highest in those assigned to the control arm. Three of the 5 metabolic syndrome components were quantified in this study (glucose $>$ 100 mg/dL, HDL-cholesterol $<$ 40 mg/dL [50 mg/dL for women] and triglycerides $>=$ 150 mg/dL). No subject met all three criteria on any study day.

Significant differences among days were seen for cholesterol measures, TG, GLU and INS (Table 2). In comparing day 14 vs. day 0, exposure to efavirenz and amprenavir significantly increased TC (least-squares estimate [LSE] of change was 15 mg/dL), LDL-c (10 mg/dL), HDL-c (2.5 mg/dL), TG (16 mg/dL), and GLU (2.3 mg/dL). TCHC and INS changes did not differ from zero. An additional 7 days of exposure to efavirenz plus amprenavir (day 21 vs. day 14), with or without a second PI (pooling arms), increased TC (LSE change from day 14, 13 mg/dL), LDL- (9 mg/dL), TCHC (0.37) and TG (18 mg/dL). HDL-c, GLU, and INS were not changed further from day 14. Overall, comparing day 42 vs. day 0, ARV exposure was discontinued, but TC remained significantly elevated over baseline (29 mg/dL), as did LDL-c (20 mg/dL), HDL-c (7.0 mg/dL), and INS (1.0 μU/mL). TCHC, TG, and GLU values, however, were restored to near baseline at this follow-up visit. Day 42 changes were not associated with length of time since drug discontinuation (actual range 1-5 weeks).

On day 21, only TCHC differed with second PI. TCHC increases on the nelfinavir and control arms were larger than increases on the other arms. HOMA and CPEP values did not differ by day, nor by second PIs.

Higher amprenavir AUCs on day 14 were associated with larger TCHC, TG, and CPEP elevations above baseline (Table 3). A regression equation was derived to predict TG increments for a given amprenavir AUC on day 14: [27.45 − (0.34 × baseline TG) + (5.12 × amprenavir AUC)]. For a subject with baseline TG = 79.5 mg/dL (median), an amprenavir AUC of 4.0 or 8.0 μg/mL would be predicted to increase TG by 21 or 41 mg/dL, respectively. Larger amprenavir AUCs also predicted smaller increases in HDL-c on day 14. On day 14, larger efavirenz AUCs were associated with larger HDL-c increases and smaller TG increases.

DISCUSSION

In HIV-seronegative subjects, short-term exposure to the NNRTI efavirenz plus the PI amprenavir increased total- and LDL-cholesterol levels 10% above baseline. Subsequent exposure to a second PI (saquinavir, nelfinavir, indinavir, or ritonavir) further increased total-

TABLE 1. SUMMARY STATISTICS OF METABOLIC PARAMETERS ON EACH STUDY DAY (ALL ARMS POOLED)

Fasting parameters	N	*Day*	*Mean*	*SD*	*Range*	*% with NCEP Metabolic syndrome criteria*
Total cholesterol	51	0	155.5	25.1	110.0–200.0	0.0%
(mg/dL)	51	14	170.3	28.1	95.0–226.0	0.0%
	49	21	183.2	33.0	127.0–245.0	6.1%
	40	42	186.2	35.1	107.0–279.0	5.0%
LDL-cholesterol	51	0	90.4	25.9	33.8–149.2	0.0%
(calc) (mg/dL)	51	14	100.0	25.4	40.8–158.2	0.0%
	47	21	109.0	30.6	50.6–177.2	4.3%
	39	42	111.6	34.2	43.8–214.0	7.7%
HDL-cholesterol	51	0	45.3	12.7	19.0–80.0	31.3%
(mg/dL)	51	14	47.8	12.5	28.0–79.0	27.5%
	49	21	47.1	1.5	27.0–74.0	28.6%
	40	42	51.8	13.8	31.0–87.0	17.5%
Non-HDL-	51	0	110.2	23.7	58–164	0.0%
cholesterol	51	14	122.5	26.7	58–175	0.0%
(mg/dL)	49	21	136.1	32.8	71–212	4.1%
	39	42	134.4	34.5	67–229	5.1%
Total:HDL	51	0	3.64	0.97	2.02–5.89	—
cholesterol	51	14	3.74	0.96	2.25–5.77	—
ratio	49	21	4.11	1.22	2.27–8.31	—
	39	42	3.80	1.05	2.23–6.29	—
Triglycerides	50	0	93.9	46.7	45.0–221.0	18.0%
(mg/dL)	50	14	109.7	50.2	50.0–244.0	16.0%
	47	21	123.0	57.3	50.0–332.0	23.4%
	40	42	106.8	57.1	40.0–288.0	15.0%
Glucose (mg/dL)	52	0	89.3	7.9	76.0–115.0	7.7%
	52	14	91.5	7.5	71.0–110.5	9.6%
	51	21	91.1	5.7	79.0–101.0	2.0%
	43	42	89.2	5.7	74.0–101.5	2.3%
Insulin (μU/mL)	50	0	6.73	3.01	2.00–16.10	2.0%
	50	14	6.45	3.48	1.10–20.60	2.0%
	47	21	6.66	3.37	2.00–15.40	2.1%
	40	42	7.68	3.83	2.00–19.20	5.0%
Insulin resistance	50	0	1.51	0.74	0.42–4.05	—
(HOMA)	50	14	1.47	0.83	0.26–4.41	—
	47	21	1.51	0.80	0.42–3.66	—
	40	42	1.71	0.90	0.38–4.47	—
C-peptide	52	0	1.61	0.65	0.61–3.80	—
(ng/mL)	52	14	1.53	0.70	0.46–3.30	—
	50	21	1.59	0.71	0.54–3.90	—
	41	42	1.59	0.75	0.60–3.70	—

NCEP ATP III threshold levels: total cholesterol ≥240 mg/dL, LDL-c ≥160 mg/dL, HDL-c <40 (men) <50 (women) mg/dL, non-HDL-c >190 mg/dL, TG ≥150, glucose ≥100 mg/dL, insulin ≥15 μU/mL.

and LDL-cholesterol levels (~20% above baseline). Several healthy normal subjects developed components of the cardiometabolic syndrome: 29% of men had HDL-c <40 mg/dL, 23% of subjects had TG > 150 mg/dL, and 6% of subjects had TC >240 mg/dL on day 21. (HDL-c was above 50 mg/dL for the one woman with available data.) Of note, HDL-c increased after 14 days of efavirenz/amprenavir, but unlike TC, did not rise further after introduction of a second PI; hence TCHC was elevated only on day 21. Total-, LDL-, and HDL-c remained elevated above baseline 1–5 weeks after discontinuing all medications. These findings confirm that, even in healthy, normolipidemic people not infected with HIV, short-term exposure to efavirenz plus one and two PIs can rapidly induce alterations in fasting lipid and

TABLE 2. ALTERATIONS IN LIPID/LIPOPROTEIN AND GLUCOREGULATORY PARAMETERS AFTER EFAVIRENZ PLUS AMPRENAVIR (DAY 14 VS. DAY 0), AFTER CONTINUED EFAVIRENZ AND AMPRENAVIR, PLUS 7 DAYS OF A SECOND PI (DAY 21 VS. 14), AND AFTER DISCONTINUING ALL ARV MEDICATIONS (DAY 42 VS. 0)

	Day 14 vs. 0				*Day 21 vs. 14*				*Day 42 vs. 0*			
Measure	N	*LSM*[a]	*SE*[b]	*95% CI*[c]	N	*LSM*	*SE*	*95% CI*	N	*LSM*	*SE*	*95% CI*
T-C	51	**14.8**	3.8	**7.4, 22.3**	49	**13.1**	3.8	**5.6, 20.7**	40	**29.1**	4.1	**21.0, 37.2**
LDL-C	51	**9.6**	3.5	**2.8, 16.4**	47	**8.7**	3.6	**1.6, 15.7**	39	**20.1**	3.8	**12.6, 27.6**
HDL-C	51	**2.5**	1.0	**0.4, 4.6**	49	−0.8	1.1	−2.9, 1.3	40	**6.8**	1.1	**4.6, 9.1**
TCHC	51	0.10	0.09	−0.08, 0.28	49	**0.37**	0.09	**0.19, 0.55**	39	0.06	0.10	−0.14, 0.26
TG	50	**15.8**	7.5	**0.9, 30.6**	47	**17.7**	7.7	**2.6, 32.9**	40	9.5	8.1	−6.4, 25.5
GLU	52	**2.26**	1.04	**0.20, 4.32**	51	−0.42	1.05	−2.49, 1.65	43	−0.29	1.11	−2.47, 1.90
INS	50	−0.28	0.42	−1.11, 0.55	47	0.17	0.43	−0.68, 1.02	40	**1.04**	0.45	**0.14, 1.93**

[a]Least squares mean of change; bolded values are significant (95% CI does not include the value zero).
[b]Associated standard error.
[c]95% confidence interval around least-square mean change; bolded if significant.
All units are mg/dL, except insulin (μU/mL).

lipoprotein parameters in the absence of weight gain. Depending on the magnitude of the NNRTI plus PI-induced increases in TG, TC, LDL-c, HDL-c, the cumulative effects on lipid/lipoprotein parameters may be pro-atherogenic, and indicate that caution should be exercised when dual PI-containing regimens (e.g., ritonavir boosting) are prescribed.

The lipid/lipoprotein alterations observed here are consistent with earlier reports in uninfected volunteers showing 10-40% increases in total- and HDL-cholesterol following short-term exposure to efavirenz with NRTIs or indinavir.[42] They are also consistent with the short-term metabolic effects of amprenavir-based antiretroviral regimens on fasting serum

TABLE 3. PREDICTED CHANGES IN METABOLIC PARAMETERS BASED ON OBSERVED AMPRENAVIR AND EFAVIRENZ AUCS

Amprenavir AUC in plasma (μg · h/mL)[a]	*Predicted change in metabolic parameter (associated 95% confidence intervals)*				
	HDL-c, day 14[b]	*TCHC, day 14*[c]	*TG, day 14*[d]	*GLU, day 21*[e]	*CPEP, day 14*[f]
0.5	7.44	−0.17	3.04	1.87	−0.38
4.0	3.10	0.12	20.96	1.25	−0.04
8.0	−1.86	0.46	41.45	0.54	0.34
14.0	−9.29	0.96	72.18	−0.52	0.92
Efavirenz AUC in plasma (μg · h/mL)					
20.0	0.64	—	35.78	—	
38.0	2.29	—	24.79	—	
170.0	14.34	—	−55.83	—	

[a]Representative plasma amprenavir AUCs were derived from A5043 as follows: 0.5 was the minimum AUC, 4.0 represents the median AUC on days 14 and 21, 8.0 represents the median AUC on day 0, and 14.0 represents the maximum AUC on day 14. For efavirenz AUCs: 20.0, 38.0 and 170.0 represent the minimum, median and maximum values obtained on A5043 subjects.

[b]HDL-cholesterol, mg/dL. The estimated regression equations were: (1) for changes on day 14, 19.43—(0.26*baseline HDL–c)—1.24 (0.97*day 14 amprenavir AUC), and (2) for additional changes on day 21, 8.81—(0.23*baseline HDL-c) + (0.09* day 14 efavirenz AUC). In generating predicted values, baseline median HDL-c (43.0) was used. (Baseline values were predictive of day 14 changes from baseline.)

[c]Ratio of total:HDL-cholesterol, change from baseline on day 14. The estimated regression equation was: 0.57—(0.22*baseline TCHC) + (0.08*day 14 amprenavir AUC). Baseline median TCHC (3.57) was used.

[d]Triglycerides, change from baseline on day 14, mg/dL. The estimated regression equations were: (1) as a function of amprenavir AUCs, 27.45—(0.34*baseline TG) + (5.12*day 14 amprenavir AUC), and (2) as a function of efavirenz AUCs, 68.76—(0.26*baseline TG)—(0.61*day 14 efavirenz AUC). Baseline median TG (79.5) was used.

[e]Blood glucose, change from day 14 on day 21, mg/dL. The estimated regression equation was: 1.95—(0.18*day 14 amprenavir AUC). Baseline GLU, not a significant predictor, was not included as a model covariate.

[f]C-peptide, change from baseline on day 14, ng/mL. The estimated regression equation was: 0.09—(0.34*baseline CPEP) + (0.10*day 14 amprenavir AUC). Baseline median CPEP (1.5) was used.

lipid/lipoprotein levels, with modest effects on glucose and insulin levels, in HIV-infected people,[43] and with long-term metabolic effects of efavirenz-based regimens on fasting serum lipid/lipoprotein levels in HIV-infected people.[44] In these latter two studies, 48 weeks of antiretroviral therapy was associated with weight and fat gain, and these may have contributed to the development of dyslipidemia or insulin resistance. In the present study, weight changes were minimal and could not account for the metabolic alterations observed, suggesting that intrinsic pharmacokinetic-pharmacodynamic effects mediated the metabolic alterations.

On day 14, when all subjects were exposed to efavirenz plus amprenavir, higher plasma amprenavir AUCs were associated with larger increases in triglycerides, C-peptide and TCHC ratio. In contrast, subjects with larger amprenavir AUCs tended to have smaller increases in HDL-cholesterol (day 14) and in glucose (day 21). For a hypothetical subject with a baseline triglyceride of 79.5 mg/dL and an amprenavir AUC of 14.0 μg/mL, our regression model predicted a triglyceride increase of 72.2 mg/dL, resulting in an expected triglyceride level of 152 mg/dL, which exceeds the NCEP classification (TG >150mg/dL). These findings strongly suggest that pharmokinetic interactions among ARV medications, and the resulting 'boosted' levels of certain ARV medications, in this case amprenavir, can adversely alter lipid/lipoprotein levels. The findings further suggest that ARV pharmacokinetic-pharmacodynamic interactions can occur rapidly, can dysregulate lipid/lipoprotein, and to a lesser extent, glucose metabolism, can occur at subtherapeutic doses of amprenavir, and may augment the risk for cardiometabolic syndrome.

Significant elevations in total-, HDL-, and LDL-cholesterol and insulin persisted 2–3 weeks after drug exposure, whereas triglycerides and glucose returned to baseline levels more quickly. The magnitude of elevation was not associated with time since drug discontinuation. Triglyceride, but not total-cholesterol, levels have been reported to return to baseline in some but not all PI-to-NNRTI switch studies. Augmented cholesterol and lipoprotein production rates, along with impairments in cholesterol and lipoprotein clearance rates, are responsible for dyslipidemia in HIV-infected people, both before and after initiating ARV therapy.[45,46] Whether the persistent lipid/lipoprotein changes noted here were due to ARV pharmokinetic-pharmacodynamic interactions and their effects on cholesterol-lipoprotein production and/or clearance rates is not clear, but requires further investigation.

The largest cholesterol and triglycerides changes from baseline were seen on day 21, when subjects were taking efavirenz, amprenavir, and a second PI (in 4 of 5 arms). Potential explanations for the additional alterations (day 21 vs. 14) in lipid/lipoprotein parameters include (1) extended duration of efavirenz and amprenavir exposure, (2) direct effects of the added PI, or (3) increased amprenavir levels (on some arms) secondary to the added PI. The present study was neither designed nor powered to differentiate among these factors. In particular, if direct effects due to second PIs truly existed, it appears that they were either obscured by arm differences in amprenavir levels or undetectable due to small sample sizes. It is interesting that although amprenavir AUCs exhibited much wider variation on day 21, more of the day 14 metabolic changes were associated with amprenavir levels.

In this short-term study, a prolonged pharmacologic effect may have induced sustained biochemical alterations. Alternately, prolonged elimination of efavirenz may have contributed to the metabolic changes observed. Clinically and statistically significant changes on day 14 seem more likely due to 13 days of efavirenz rather than 3 days of amprenavir dosing. The longer half-life of efavirenz ($\geq$50h)[42] relative to the other drugs suggests that it may have a role in the persistence of metabolic alterations 2–3 weeks after simultaneous discontinuation of all study drugs. The short-term effects of amprenavir on lipids and metabolic parameters demonstrated during concurrent efavirenz administration should be viewed in the context of long-term studies of amprenavir without efavirenz given. In one study of 455 HIV-infected people,[47] 48 wks of amprenavir use was associated with severe hypertriglyceridemia (>750 mg/dL), hyperglycemia (>250 mg/dL),

or hypercholesterolemia (>320 mg/dL) in only 5%, 1%, and <1% of the participants, respectively. In a review of amprenavir safety data from adult study treatment groups, median values for triglyceride, glucose, and total-cholesterol levels showed no clinically significant changes over 64 weeks, although slight initial increases in median total-cholesterol levels were observed.[48] In PRO3001, the incidence of grade 3–4 increases in TG was 0% with amprenavir/lamivudine/zidovudine (n = 111) and <1% with lamivudine/zidovudine (n = 108).

There were some limitations. Healthy HIV-seronegative men and women were enrolled to examine the effects of select ARV medication exposures without the potential confounders of HIV-infection, comorbidities, and other ARV drug classes. Thus, the complex clinical condition typical of HIV-1 infected patients is oversimplified. Efavirenz and protease inhibitor pharmacokinetic parameters were similar to those that have been reported in HIV-infected patients, and our findings provide a framework for interpreting ARV-induced metabolic changes in HIV-infected people. Currently, ritonavir-boosted ARV regimens are used most commonly. These were not a focus of the current study (which was designed several years ago). Future studies will examine the metabolic alterations that occur with newer PIs (e.g., lopinavir, atazanavir) boosted by ritonavir.

CONCLUSION

In conclusion, pharmacokinetic and pharmacodynamic drug interactions resulting when efavirenz was combined with single or dual PIs are complex, should be examined with regard to individual medications, and may induce elevations in serum lipid/lipoprotein levels that are proatherogenic. Changes in serum lipids noted during short-term treatment did not return to baseline after drug discontinuation, suggesting that a prolonged pharmacologic effect—possibly sustained, direct pharmacologic action or delayed efavirenz elimination—needs to be examined further with dual-PI regimens that include ritonavir pharmacokinetic enhancement, with and without efavirenz.

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