Impact of steady-state lopinavir plasma levels on plasma lipids and body composition after 24 weeks of lopinavir/ritonavir-containing therapy free of thymidine analogues

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Objectives: To study the impact of lopinavir/ritonavir-containing therapy on plasma lipids and body fat of HIV-infected adults and to assess whether lopinavir plasma levels at steady state are correlated with plasma lipids and body fat after 24 weeks.

Methods: Patients had their antiretroviral therapy switched to an antiretroviral regimen containing lopinavir/ritonavir plus one or two non-thymidine analogues. Body composition was assessed by dual energy X-ray absorptiometry at baseline and at week 24 and an intensive pharmacokinetic (PK) 12 h profile was performed at week 2.

Results: Twenty-six patients were included. Plasma triglycerides (from 206 mg/dL to 261 mg/dL, P = 0.09) and total cholesterol (from 201 to 206 mg/dL, P = 0.03) increased from baseline to week 24. There was a significant rise in total fat (from 10.9 to 11.9 kg, P = 0.02) and limb fat (from 3.8 to 4.4 kg, P = 0.02) from baseline to week 24. We did not find any correlation between PK lopinavir levels and changes over time for triglycerides, cholesterol or body fat composition.

Conclusions: There was an increase in plasma triglycerides and total cholesterol levels and a gain in both total and limb fat at 24 weeks, but these changes were not correlated with lopinavir plasma levels.

Keywords: pharmacokinetic parameters, body composition, hypertriglyceridaemia, hypercholesterolaemia

Introduction

Lipoatrophy and dyslipidaemia are common events in HIV-infected patients.1–4 Influence of a treatment with a combination of antiretroviral therapy for HIV-1 infection with protease inhibitors (PIs), nucleoside analogue reverse transcriptase inhibitors (NRTIs), or both, on the development of metabolic and lipid disturbances has been well established,5 particularly in the case of thymidine analogues ( stavudine and zidovudine) and PIs boosted with ritonavir.

Kaletra® (soft-gel capsules), a fixed-dose co-formulation of lopinavir and ritonavir, has demonstrated a potent antiviral activity in antiretroviral-naive and -experienced patients. However, patients who initiate lopinavir/ritonavir-containing antiretroviral therapy may show an increased incidence of hyperlipidaemia.6–8 Recently, a new formulation of lopinavir/ritonavir as a tablet formulation, Kaletra Meltrex®, is available. Several studies are underway to determine if this new formulation of lopinavir has a different metabolic behaviour. To date, the available metabolic information on lopinavir/ritonavir is related to the soft-gel capsules.

Among several factors contributing to a higher risk of clinically significant metabolic abnormalities in patients receiving lopinavir/ritonavir,8–10 it has been hypothesized that the higher the plasma levels of lopinavir, the higher the risk of hypertriglyceridaemia and hypercholesterolaemia.

The impact of lopinavir/ritonavir on body fat is scarcely known. It has been reported that switching from stavudine or zidovudine to an NRTI-sparing regimen containing lopinavir/ritonavir plus nevirapine or efavirenz was associated with a significant improvement in subcutaneous abdominal adipose tissue,11 and recently, a study has been published where

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antiretroviral-naïve patients were randomized to lopinavir/ritonavir plus saquinavir or zidovudine/lamivudine and where, according to our results, a statistically significant increase in the lower extremities fat has been shown.\textsuperscript{12}

It has been suggested that changes in body fat composition in patients treated with lopinavir/ritonavir might also be related to lopinavir plasma levels.\textsuperscript{13} Some studies have found significant correlations between lopinavir trough concentrations and changes in plasma lipid profile\textsuperscript{14,15} but not others.\textsuperscript{16,17} However, in all these studies, changes in lipids or in fat values cannot be reliably interpreted because other potential confounding factors (e.g. concurrent thymidine analogue use, baseline plasma lipids or hepatitis C virus co-infection) were not considered.

We aimed to study fasting plasma lipids and body fat after 24 weeks of an antiretroviral regimen containing lopinavir/ritonavir free of thymidine nucleosides and to assess whether there was any correlation between changes in plasma lipids and body fat and lopinavir plasma concentration.

Methods

Patients

HIV-infected antiretroviral-experienced adults naive for lopinavir/ritonavir were eligible. Exclusion criteria were pregnancy, concomitant therapies with investigational drugs, steroids and agents known to have major interactions with lopinavir/ritonavir. Lopinavir/ritonavir was dosed at 400/100 mg twice daily in all participants.\textsuperscript{18} Concomitant therapy with one or two non-thymidine nucleosides was allowed according to physician discretion.

Patients receiving stable anti-diabetic or lipid-lowering therapies before the study could be included, but no changes in dosing and no new prescriptions of these drugs were allowed during the study. No specific interventions to modify diet or physical exercise were done. The local Human Research Ethics Committee approved the study. All patients provided written informed consent. Patients were evaluated at baseline and at least at 12 and 24 weeks thereafter.

Laboratory parameters

At each visit, clinical and fasting laboratory data including at least CD4 T lymphocyte count and HIV-1 RNA, glucose, triglycerides, total cholesterol, high-density lipoprotein (HDL) cholesterol and low-density lipoprotein (LDL) cholesterol levels were obtained. Laboratory parameters were measured as described elsewhere.\textsuperscript{8} The cut-off points for defining clinically significant triglyceride and cholesterol levels were chosen according to the National Cholesterol Education Program guidelines.\textsuperscript{19}

Body composition parameters

Clinically evident lipodystrophy at baseline was defined as the presence of any body fat loss that was clinically evident for both the patient and the physician as previously described.\textsuperscript{8} Total and regional fat were quantified with dual energy X-ray absorptiometry (DEXA; Lunar Prodigy, Madison, WI, USA) at baseline and at week 24.\textsuperscript{20}

Pharmacokinetic parameters

Plasma concentrations of lopinavir were analysed by a modified validated HPLC assay developed in our Clinical Pharmacology Laboratory,\textsuperscript{21} between week 2 and week 3 after lopinavir/ritonavir treatment initiation to ensure steady-state lopinavir levels\textsuperscript{22} and repeated at week 24.

An intensive pharmacokinetic (PK) 12 h dosing interval study was performed including lopinavir trough plasma concentrations (defined as pre-dose concentration 12 h after the lopinavir/ritonavir dinner dose) and concentrations at 1, 2, 3, 4, 5, 6, 8 and 12 h after the lopinavir/ritonavir morning dose\textsuperscript{23,24} plus a standard breakfast (15 mg of bread, 25 g of butter, 34 g of jam and 100 mL of milk) that were measured in the Clinical Pharmacology Unit.

The AUC\textsubscript{12} (area under the drug plasma concentration–time curve over 12 h), C\textsubscript{max} (peak plasma drug concentration) and C\textsubscript{trough} (trough plasma concentration) were determined by non-compartmental methods,\textsuperscript{25} using the WinNonlin Pro pharmacokinetic program (version 3.1; Scientific Consulting Inc., Cary, NC, USA).

Statistical analysis

A sample size of 25 patients was considered adequate to detect a mean increase in plasma triglycerides of at least 25 mg/dL with 80% power and an alpha error of 0.05, according to previous data from our clinics.\textsuperscript{8} Variables for the analysis were chosen because of their potential impact on plasma lipids and body fat, and included sex, age, weight, height, body mass index (BMI), route of HIV transmission, alcohol consumption (defined by regular daily intake of alcoholic beverages), diabetes mellitus (defined by clinical diagnosis), hepatitis C virus infection (defined by serological diagnosis), clinically evident lipodystrophy, use of PIs prior to lopinavir/ritonavir (defined as regular intake of those drugs for ≥1 month), use of lipid-lowering therapy at baseline (defined as regular intake for ≥1 month), antiretroviral drugs considered within the regimen containing lopinavir/ritonavir, virological suppression (defined as regular intake of at least 200 copies/mL) and fasting plasma values of glucose, triglycerides, total cholesterol, HDL cholesterol and LDL cholesterol at baseline before the initiation of lopinavir/ritonavir and 12 and 24 weeks after.

Quantitative characteristics were described with median and interquartile range (IQR) and qualitative characteristics with frequencies and percentages. Percentage of change over time for continuous variables was defined as the difference between the value at week 24 and the value at baseline, adjusted for the latter.

Comparisons of laboratory and body composition parameters at each time point were performed using a Wilcoxon signed-rank test. The correlation between the PK parameters at baseline and the changes in lipid profile (absolute values or categorized as previously defined PK\textsuperscript{15} or ATPIII cut-offs\textsuperscript{19}) or body fat composition (absolute values or categorized under or above 20% of total body fat\textsuperscript{26}) were evaluated by the Spearman’s rank coefficient.

Variability between individuals (inter-individual variability) and within individuals (intra-individual variability) was calculated using the coefficient of variation expressed as a percentage.

A median regression model was carried out to identify predicting factors for developing a clinically significant high plasma level of glucose (≥126 mg/dL), triglycerides (≥200 mg/dL) and cholesterol (≥200 mg/dL) and an increase in body fat compared with baseline. The following variables were included: age, sex, weight, BMI, risk behaviour, AIDS-defining illnesses, hepatitis C virus, previous clinically evident lipodystrophy, previous dyslipidaemia according to ATPIII,\textsuperscript{19} lack of experience of PIs, withdrawal of nucleosides, non-nucleosides or PIs on entering this study, concomitant antiretrovirals used, baseline CD4, baseline viral load, high cholesterol baseline levels (≥200 mg/dL), high triglyceride baseline levels (≥200 mg/dL), high cholesterol, LDL or HDL baseline levels (≥130 and
60 mg/dL respectively), limb fat percentage at baseline, trunk fat percentage at baseline, total body fat percentage at baseline, lopinavir C_{\text{AUC}} and lopinavir C_{\text{max}} at baseline. Those variables with a $P < 0.1$ in a univariate analysis were entered into a multivariate analysis through a stepwise selection process.

Statistical analyses were performed using STATA (Stata Statistical Software: Release 8.0, 2003; Stata Corporation, College Station, TX, USA).

## Results

### Patients

Thirty-eight patients were eligible. Of them, 12 (31.5%) were excluded from the final analyses, because of one of the following reasons: never started assigned antiretroviral therapy ($n = 2$, 5.3%), were lost to follow-up ($n = 6$, 15.8%), discontinued lopinavir/ritonavir due to diarrhea ($n = 3$, 7.9%) or withdrawn consent ($n = 1$, 2.6%). Baseline characteristics of the 26 patients who completed the study are summarized in Table 1.

Concomitant antiretrovirals associated with lopinavir/ritonavir were: didanosine in 12 (46.2%); tenofovir in 16 (61.5%); lamivudine in 9 (34.6%); abacavir in 2 (7.7%); nevirapine in 3 (11.5%) and efavirenz in 6 (23.1%). Antiretroviral therapy before the study had consisted of three nucleoside analogues in 7 patients (26.9%), two nucleosides plus a non-nucleoside analogue in 11 patients (42.3%) and two nucleosides plus a PI in 9 patients (34.6%). Ten and six patients (61.5%) had left zidovudine and stavudine, respectively, at study entry (Table 1).

At baseline, no patient had been previously diagnosed with diabetes and none of them had a significant alcohol consumption (>40 g/day). Three (11.5%) patients were receiving lipid-lowering therapy but it was not modified during follow-up. No patient developed any cardiovascular event during the study period.

### Laboratory parameters

Median and 25th and 75th percentile plasma levels of triglycerides and total, HDL and LDL cholesterol at baseline and at week 24 are represented in Figure 1(a).

Triglycerides showed a trend to increase ($P = 0.09$) from baseline (median 206 mg/dL, IQR 124–257 mg/dL) to week 24 (median 261 mg/dL, IQR 173–336 mg/dL). Eleven patients (42.3%) changed from a normal level of triglycerides (median 261 mg/dL, IQR 173–336 mg/dL). Eleven patients (42.3%) changed from a desirable level of total cholesterol (less than 200 mg/dL) to week 24. Protective factors for triglycerides $>200$ mg/dL at week 24 in the multivariate median regression model were a concomitant treatment with tenofovir [adjusted odds ratio (AOR) 0.03; CI 0.001–0.9, $P = 0.04$] and baseline lip lume AUC (AOR 0.7; CI 0.5–0.9, $P = 0.04$) (to have more limes fat at baseline is associated with a lack of increase in fat using the study medication).

Total cholesterol increased significantly ($P = 0.03$) from baseline (median 201 mg/dL, IQR 171–241 mg/dL) to week 24 (median 206 mg/dL, IQR 187–228 mg/dL). Three patients (12%) changed from a desirable level of total cholesterol (less than 200 mg/dL) at baseline to a borderline high level (200–239 mg/dL) at week 24, whereas the remaining patients kept their baseline cholesterol levels within desirable range (Table 1).

### Table 1. Baseline characteristics of the patients

| Number | Age (years) | Number of males/females | Body weight (kg) | BMI (kg/m²) | HIV risk group (number/%) | Number of previous undetectable viral loads (HIV-1 RNA <200 copies/mL) | Number with a prior AIDS-defining event | Prior NRTIs (number, % of patients) | Clinically evident lipodystrophy | Log_{10} HIV-1 RNA (copies/mL) | CD4 + T cell count (cells/mm³) |
|--------|-------------|-------------------------|------------------|-------------|--------------------------|---------------------------------------------------------------|----------------------------------------|----------------------------------|---------------------------------|-------------------------------|--------------------------------|-----------------------------|
| 26     | 43 (IQR 40–51) | 22/4                    | 64 (IQR 60–72)   | 22.3 (IQR 21.2–24.0) | homossexuals or bisexuals 10 (38.5%) | 11 (34.6%)                                                   | 12 (46.2%)                             | zidovudine 10 (38.4%)                | 9 (34.6%)                       | 3.1 (IQR 2.3–4.3)             | 344 (IQR 113–499)            |
|        |             |                         |                  |             | heterosexuals 6 (23.1%)  |                                |                                                       | stavudine 6 (23.1%)                    | 12 (46.2%)                       |                                |                                |                        |
|        |             |                         |                  |             | intravenous drug users 10 (38.5%) |                                |                                                       | lamivudine 16 (61.5%)                  |                                |                                |                                |                        |
|        |             |                         |                  |             | Number with a prior AIDS-defining event 12 (46.2%) |                                |                                                       | didanosine 12 (46.2%)                 |                                |                                |                                |                        |
|        |             |                         |                  |             | Number of previous HAART regimens 3.5 (IQR 2–6) |                                |                                                       | abacavir 8 (30.7%)                     |                                |                                |                                |                        |
|        |             |                         |                  |             | Tenofovir 5 (19.2%) |                                |                                                       | tenofovir 5 (19.2%)                   |                                |                                |                                |                        |
|        |             |                         |                  |             | Total cholesterol (mg/dL), n = 26 |                                |                                                       |                          |                                |                                |                                |                        |
|        |             |                         |                  |             | <150 4 (15.4%) |                                |                                                       |                          |                                |                                |                                |                        |
|        |             |                         |                  |             | 150–199 6 (23.1%) |                                |                                                       |                          |                                |                                |                                |                        |
|        |             |                         |                  |             | 200–499 12 (46.1%) |                                |                                                       |                          |                                |                                |                                |                        |
|        |             |                         |                  |             | ≥500 4 (15.4%) |                                |                                                       |                          |                                |                                |                                |                        |
|        |             |                         |                  |             | LDL cholesterol (mg/dL), n = 22 |                                |                                                       |                          |                                |                                |                                |                        |
|        |             |                         |                  |             | <100 9 (34.6%) |                                |                                                       |                          |                                |                                |                                |                        |
|        |             |                         |                  |             | 100–159 8 (30.7%) |                                |                                                       |                          |                                |                                |                                |                        |
|        |             |                         |                  |             | >160 5 (19.2%) |                                |                                                       |                          |                                |                                |                                |                        |
|        |             |                         |                  |             | Fat (kg), n = 24 |                                |                                                       |                          |                                |                                |                                |                        |
|        |             |                         |                  |             | total fat 11.0 (IQR 9.6–14.5) |                                |                                                       |                          |                                |                                |                                |                        |
|        |             |                         |                  |             | trunk fat 6.5 (IQR 5.0–8.4) |                                |                                                       |                          |                                |                                |                                |                        |
|        |             |                         |                  |             | limb fat 3.8 (IQR 2.8–5.4) |                                |                                                       |                          |                                |                                |                                |                        |

Values are expressed as median (IQR) or number (%) unless otherwise stated. LPV/r, lopinavir/ritonavir.

In a multivariate median regression model, clinically evident lipodystrophy (AOR 21.2; CI 1.1–396.3, $P = 0.04$) and cholesterol $>200$ mg/dL (AOR 24.3; CI 1.9–307.2, $P = 0.02$) at baseline were identified as independent factors predicting total cholesterol $>200$ mg/dL at week 24.

In contrast, neither glucose nor HDL and LDL cholesterol changed significantly from baseline (median 86 mg/dL, IQR 77–97 mg/dL; median 45 mg/dL, IQR 36–55 mg/dL; median 107 mg/dL, IQR 90–136 mg/dL, respectively) to week 24 (median 89 mg/dL, IQR 82–97 mg/dL; median 42 mg/dL, IQR 36–53 mg/dL; median 106 mg/dL, IQR 98–126 mg/dL) ($P = 0.6; P = 0.5; P = 0.4$, respectively).
**Effect of lopinavir levels on lipids and fat**

Figure 1. (a) Changes in metabolic parameters over time. Baseline, plasma levels of metabolic parameters before initiating a lopinavir/ritonavir-based treatment; W24, plasma level of metabolic parameters after 24 weeks of a lopinavir/ritonavir-containing combination antiretroviral therapy; TRIGLY., triglycerides; T. COL, total cholesterol; LDL COL, low-density lipoprotein cholesterol; HDL COL, high-density lipoprotein cholesterol. P values calculated through Wilcoxon signed-rank test. (b) Changes in body fat over time. Baseline, body fat composition before starting a lopinavir/ritonavir-based antiretroviral therapy; W24, body fat composition after 24 weeks of a lopinavir/ritonavir-based antiretroviral therapy. P values calculated through Wilcoxon signed-rank test.

**Body composition**

Median values and IQR of DEXA body fat percentage (total, limbs and trunk) at baseline and at week 24 are summarized in Figure 1(b).

Total body fat increased significantly from baseline to week 24 (from a median of 10.9 kg, IQR 9.5–14.6 kg, to a median of 11.9 kg, IQR 8.7–18.1 kg). There was a median increase in the percentage of the body fat of 1.5% by week 24 (from a median body fat of 17.2%, IQR 15.0–25.0%, to a median of 18.7%, IQR 16.3–26.3%; P = 0.009).

Trunk body fat increased significantly from baseline to week 24 (from a median of 6.5 kg, IQR 5–8.7 kg, to a median of 7.3 kg, IQR 4.5–11.1 kg). There was a median increase in the percentage of the trunk body fat of 1.25% by week 24 (from a median trunk fat of 22.7%, IQR 16.6–28.8%, to a median of 24.0%, IQR 18.5–32.6%; P = 0.03).

Limb body fat increased significantly from baseline to week 24 (from a median of 3.8 kg, IQR 2.7–5.5 kg, to a median of 4.4 kg, IQR 3.1–6.4 kg). There was a median increase in the percentage of the limb body fat of 1% by week 24 (from a median limb fat of 30.9%, IQR 20.6–43.1%, to a median of 31.9%, IQR 22.9–44.8%; P = 0.002).

Significant correlations between fat gain in the different regions (trunk and limb) were found [Spearman’s rho (r): 0.7, P = 0.01].

In a univariate analysis, hepatitis C co-infection (crude odds ratio 0.07; CI 0.008–0.6, P = 0.02) was the only significant factor identified influencing body fat change (having a hepatitis C co-infection is associated with a lack of increase in body fat using the study medication). A multivariate analysis was not able to identify any independent factor.

**Table 2. Lopinavir plasma levels, median (IQR1–3)**

<table>
<thead>
<tr>
<th></th>
<th>Weeks 2–3</th>
<th>Week 24</th>
</tr>
</thead>
<tbody>
<tr>
<td>C&lt;sub&gt;trough&lt;/sub&gt; (mg/L)</td>
<td>6.2 (3.6–8.7) (n = 23)</td>
<td>4.6 (3.1–9.2) (n = 20)</td>
</tr>
<tr>
<td>C&lt;sub&gt;max&lt;/sub&gt; (mg/L)</td>
<td>11.0 (6.4–12.8) (n = 17)</td>
<td>10.0 (8.7–13.3) (n = 8)</td>
</tr>
<tr>
<td>AUC (mg·h/L)</td>
<td>93.6 (59.8–122.4) (n = 17)</td>
<td>79.3 (58–113.8) (n = 8)</td>
</tr>
</tbody>
</table>

**Lopinavir exposure and changes in body fat and metabolic parameters.** We performed an intensive 12 h PK profile in 17 patients at baseline and also at week 24 in eight patients. Lopinavir trough levels were available in 23 patients at baseline and in 20 at week 24. Patients who completed the follow-up of PK studies were not clinically different from those who did not complete. We found an excellent correlation between lopinavir trough levels and C<sub>max</sub> and AUC<sub>12</sub> at week 2 (r = 0.8 and 0.9, respectively, P = 0.001) and also at week 24 (r = 0.9 and 0.9, respectively, P = 0.001). The C<sub>trough</sub> at weeks 2–3 and at week 24 was significantly correlated (r = 0.5; P = 0.03). The lopinavir plasma levels are summarized in Table 2. The inter-individual and intra-individual variability of lopinavir was 44.2% and 31.2%, respectively. No significant relationship was found between C<sub>max</sub>, AUC<sub>12</sub> or lopinavir trough concentrations and the baseline total body weight, baseline BMI or gender.

**Lopinavir PK steady-state studies and metabolic parameters.** We did not find any significant correlation between lopinavir plasma concentrations (AUC<sub>12</sub>, C<sub>max</sub>, C<sub>trough</sub>) and changes over time for triglycerides, glucose or total, HDL cholesterol or LDL cholesterol serum levels, neither in absolute values nor with the predefined cut-off of lopinavir PK<sub>15</sub> and lipid profile. The correlations (r) between AUC<sub>12</sub>/C<sub>max</sub>/C<sub>trough</sub> and the change over time in absolute values (Δ of mg/dL) of triglycerides were low (r = 0.2/r = 0.3/r = 0.1, respectively) and non-statistically significant (P = 0.3/P = 0.2/P = 0.8, respectively). Correlations between AUC<sub>12</sub>/C<sub>max</sub>/C<sub>trough</sub> and the change in total cholesterol were also low (r = 0.2/r = 0.2/r = 0.0, respectively) and non-statistically significant (P = 0.4/P = 0.2/P = 0.9, respectively).

**Lopinavir PK steady-state studies and body fat composition.** We did not find any significant correlation between lopinavir plasma concentrations (AUC<sub>12</sub>, C<sub>max</sub> or C<sub>trough</sub>) and changes over time.
for total body fat, trunc fat or limb fat, neither in absolute values nor with predefined cut-off of lopinavir PK and DEXA.

The correlations (r) between AUC \(_{12}/C_{\text{max}}/C_{\text{trough}}\) and the change over time of the total body fat composition in absolute values (Δ kg of total body fat) were low (r = 0.09/r = 0.2/r = 0.2, respectively) and non-statistically significant (P = 0.85/P = 0.5/r = 0.5, respectively).

The correlations between AUC \(_{12}/C_{\text{max}}/C_{\text{trough}}\) and the change over time of the trunc body fat in absolute values (Δ kg of total body fat) were low (r = 0.4/r = 0.5/r = 0.1, respectively) and non-statistically significant (P = 0.09/P = 0.06/P = 0.6, respectively). Data for the limb fat were similar (AUC \(_{12}/C_{\text{max}}/C_{\text{trough}}\); r = 0.1/r = 0.05/r = 0.1; P = 0.6/P = 0.8/P = 0.5).

**Discussion**

Our results suggest that an increase in total cholesterol, triglycerides and body fat composition may be detected after 24 weeks of a lopinavir/ritonavir-based antiretroviral regimen free of thymidine analogues and that changes in plasma lipids and body fat at week 24 are not correlated with plasma levels of lopinavir at steady state.

In accordance with other previous reports, we detected increases in triglyceride and total cholesterol levels. After 24 weeks, 53% of our patients developed clinically significant hypertriglyceridaemia and 25% developed clinically significant hypercholesterolaemia.

Of interest are the data revealing a significant gain in weight and fat of the patients after 24 weeks of a lopinavir/ritonavir-based antiretroviral regimen not containing thymidine analogues and the homogeneous distribution of this gain both in truncal and limb fat. This might be particularly relevant in lipoatrophic HIV-infected patients or even in patients without apparent body changes because it seems not to promote the development of lipodystrophy.

To date, gain in fat had been shown switching from thymidine nucleosides to abacavir or tenofovir. It has been reported that switching from stavudine or zidovudine to a NRTI-sparing regimen containing lopinavir/ritonavir plus nevirapine or efavirenz was associated with a significant improvement in subcutaneous abdominal adipose tissue, in accordance with our results.

To our knowledge, this is the first intensive lopinavir PK 12 h dosing interval study addressing the issue of a potential impact of lopinavir/ritonavir on body fat and plasma lipids. Other studies to date have analysed only lopinavir C\(_{\text{trough}}\) levels. However, we were concerned about the difficulties in attendance for 12 h at hospital for PK tests in otherwise healthy active patients, and in addition, we found an excellent correlation between lopinavir C\(_{\text{trough}}\) level and C\(_{\text{max}}\) or AUC\(_{12}\). Therefore, with the aim of simplifying PK studies, we analysed only lopinavir trough levels in the remaining patients.

In terms of lipid toxicity, measuring total exposure to lopinavir with 12 h PK levels did not add more information than a single sample of C\(_{\text{trough}}\), as has been previously suggested. We did not find any correlation between lipid profile, body fat composition and lopinavir plasma levels. These results argue against the clinical usefulness of measuring lopinavir plasma levels for metabolic or body fat composition purposes.

In other clinical studies, also assessing lopinavir plasma levels, lipid and fat changes were measured without controlling for several variables that may act as confounding factors (thymidine analogue use in the current antiretroviral regimen, withdrawal of prior antiretroviral therapy, baseline lipid levels and hepatitis C virus co-infection). Because we controlled for these potential confounding factors in our study, we believe that our results may more accurately reflect the impact of lopinavir/ritonavir on metabolic abnormalities or body composition.

Although the most detailed prospective study combining an intensive lopinavir PK 12 h dosing interval, fasting lipid profile and studies of body composition to date, the results from this study should be viewed in the context of study limitations. The fact that this was a non-randomized study, a cohort mainly of men (there are sex-related differences in body composition) and using various antiretrovirals associated with lopinavir/ritonavir, could have introduced biased results. Nevertheless, the contribution of each antiretroviral was analysed separately in the regression model and no individual relationships with the gain of fat or lipids were detected except with tenofovir. As mentioned in the Methods section, the dose of lopinavir/ritonavir was not modified when co-administered with NNRTI because several studies have not found significant PK differences when maintaining the standard dose of lopinavir/ritonavir (400/100 mg) and because our study performed PK studies to ensure good plasma levels of lopinavir.

It could be argued that changes in fat or lipids observed in our study could be related not only to the use of lopinavir/ritonavir but also to the cessation of thymidine analogues prior to study entry (10 patients were treated with zidovudine and 6 with stavudine). Probably both factors are contributing; however, recent data of two different studies presented at CROI support our findings. The first study presented the metabolic outcome of ACTG 5142, a prospective randomized trial comparing NRTI-, PI- and NNRTI-sparing regimens for initial treatment of HIV infection in almost 800 patients. Their results showed significantly more lipodystrophy in the group taking NRTI or NNRTI than in the lopinavir/ritonavir group. The second study, a randomized trial in 155 antiretroviral-naïve patients, showed a significant sparing of peripheral lipodystrophy by HIV treatment with lopinavir/ritonavir + zidovudine/lamivudine induction followed by lopinavir/ritovar monotherapy compared with efavirenz + zidovudine/lamivudine. In both studies, the possible effect of lopinavir/ritonavir on fat could not be explained by the cessation of any medications, because the patients studied were antiretroviral-naïve.

In summary, our results suggest that an increase in plasma lipids but also in regional and total fat mass may be detected in HIV-infected antiretroviral-experienced patients after 24 weeks of lopinavir/ritonavir therapy free of thymidine analogues. Although lopinavir trough levels are a simple measurement showing an excellent correlation with other PK parameters, their values at steady state were not correlated with changes in plasma lipids and body fat. Therefore, according to our study, the measurement of lopinavir plasma levels at steady state to predict plasma lipids or body fat changes seems to be not justified.

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Effect of lopinavir levels on lipids and fat

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Transparency declarations

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