Modest but Variable Effect of Rifampin on Steady-State Plasma Pharmacokinetics of Efavirenz in Healthy African-American and Caucasian Volunteers

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Key words: Efavirenz, rifampin, drug-drug Interactions, Inter-individual variability, and pharmacogenetics.

Running title: Efavirenz and Rifampin Interaction

Word count: abstract – 243; text – 2,779.

Table: 2; Figure: 4; References: 43

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ABSTRACT

Efavirenz-based antiretroviral regimen is preferred during rifampin-containing tuberculosis therapy. However, current pharmacokinetic data is insufficient to guide optimized concurrent dosing. This study aimed to better characterize the effects of rifampin on efavirenz pharmacokinetics. Subjects were randomized to receive efavirenz 600 mg/day or efavirenz 600 mg with rifampin 600 mg/day for 8 days, with plasma samples collected for pharmacokinetic analysis over 24 hours on day 8. Treatments were then crossed-over after at least a two-week washout period and procedures repeated. Efavirenz concentrations were determined by HPLC, and pharmacokinetic parameters estimated by noncompartmental analysis. Efavirenz pharmacokinetic differences between treatment periods were evaluated by paired t-test. The coefficient of variation in efavirenz plasma AUC_{0-24h} was 50% and 56% in the absence and presence of rifampin, respectively. Of the 11 evaluable subjects (6 white, 5 black; 6 women, 5 men), the geometric mean AUC_{0-24h} ratio on:off rifampin (90% confidence interval) was 0.82 (0.72, 0.92), with individual AUC_{0-24h} ratios varying from 0.55 to 1.18. Five subjects had a 24-hour efavirenz concentration (C_{24h}) < 1000 ng/mL on rifampin. They were more likely to have received a lower dose in milligram/kilogram and to have lower efavirenz AUC_{0-24h} in the basal state. Although rifampin resulted in a modest reduction in efavirenz plasma exposure in subjects as a whole, there was high variability in response between subjects suggesting that efavirenz dose adjustment with rifampin may need to be individualized. Body weight and genetic factors will be important covariates in dosing algorithms.
INTRODUCTION

Efavirenz is an essential component of preferred antiretroviral regimens in treatment of human immunodeficiency virus (HIV) infection in patients co-infected with tuberculosis (TB) (5, 32, 33). The standard adult dose of 600 mg daily is associated with considerable inter-individual variability in plasma concentrations and clinical effects (10, 28, 38). There is even greater variability in efavirenz concentrations during coadministration with rifampin or rifampin-containing TB therapy (2, 13, 29). Efavirenz is primarily metabolized by hepatic CYP2B6, with some contributions from CYP3A4/5 (42), CYP2A6(30) and UGT2B7 enzymes (1). Rifampin was shown to be a potent inducer of CYP2B6 activity when bupropion was used as a probe of enzyme activity (22). Among healthy volunteers, rifampin caused a 26% and 20% reduction in mean efavirenz area under the curve (AUC) and maximum concentration (C_{max}), respectively (2). Among HIV/TB co-infected patients mean reductions of 24% and 25% in efavirenz AUC and C_{max}, respectively were reported with rifampin coadministration. In the above-mentioned studies, some subjects had higher efavirenz plasma exposure with rifampin (compared with off rifampin), suggesting a lack of a significant induction effect (2, 24).

While the clinical significance of the interactions between efavirenz and rifampin is not entirely understood, to offset the mean reduction efavirenz exposure with rifampin, some experts have recommended increasing efavirenz dose to 800 mg/day, especially when body weight is greater than 50 kg (5, 33). An extensive review of published literature did not find adequate evidence to support a dose increase with rifampin coadministration because of current lack of understanding of the likelihood of under- or overdosing individuals (11). Prior drug-drug interaction studies failed to evaluate genetic contributions to the variability in the induction effect of rifampin (2, 24), or performed in the setting of clinical care where patients are receiving
other antituberculous drugs in addition to rifampin or did not have an appropriate control period or comparator group (13, 20, 25, 35). To better characterize the effect of rifampin on efavirenz disposition as well as inter-individual differences in response to rifampin, we conducted a randomized two-period crossover pharmacokinetic study in African-American and Caucasian healthy volunteers.

METHODS

Study subjects

HIV-seronegative healthy volunteers, aged from 18 to 55 years old, who identified themselves as either Caucasian or African-Americans, were enrolled. Subjects were excluded if a clinically significant medical condition or laboratory abnormality was detected. The Institutional Review Board of Miriam Hospital reviewed and approved the study and all subjects signed written informed consent.

Study design

An open-label, two-period, crossover study was conducted. The subjects were randomized based on ethnicity to treatment with efavirenz 600 mg daily (period I) or efavirenz 600 mg plus rifampin 600 mg daily (Period II) on days 1 – 8. After sampling for efavirenz plasma concentrations on day 8 and at least a two-week washout period, subjects crossed-over and the procedures repeated. We enrolled equal numbers of Caucasian and African-Americans as well as males and females because of the reported ethnic and gender differences in CYP2B6 expression (21).

Pharmacokinetic sampling

Pharmacokinetic blood sampling for efavirenz concentrations was performed on day 8 of drug(s) administration. The administration of efavirenz was switched to mornings two days prior to the
pharmacokinetic sampling. The date and time of ingestion of study drugs were recorded in a diary and verified on the day of sampling. Blood samples were obtained at times 0, 0.5, 1, 1.5, 2, 3, 4, 6, 8, 10, 12 and 24 hours relative to day 8 efavirenz dosing. Blood was centrifuged at 3000g for 10 minutes and the plasma separated and stored in labeled tubes at – 70°C until time of drug assay.

**Pharmacokinetic analysis**

Efavirenz concentrations in plasma were measured using high-performance liquid chromatography (HPLC) with ultraviolet light method (36). This method is validated over a range of 15 to 10,000 ng/mL and is accurate (90.4 – 110.5%) with intraday and inter-day precision of 2.3 – 8.3%. The assay is validated according to FDA guidelines and the Laboratory is CLIA certified and participates in quarterly national and international external proficiency testing. Maximum efavirenz plasma concentration (C\(_{\text{max}}\)), time to C\(_{\text{max}}\) (T\(_{\text{max}}\)), and concentration at 24 hours (C\(_{24\text{h}}\)) were obtained by visual inspection of the plasma efavirenz concentration-time profile for each patient. The 24-hour post dose sample for one subject during the rifampin-dosing phase was lost during centrifugation. Since PK sampling was performed at steady state, efavirenz concentration obtained at time = 0 (907.3 ng/mL) was considered as C\(_{24\text{h}}\) for this subject (ID: W6). Noncompartmental analysis was conducted using Win Nonlin Professional v 5.2.1 (Pharsight Corporation, Cary, NC). Area under the concentration-time curve from time zero to 24 hours (AUC\(_{0-24\text{h}}\)) was calculated using log\(_{e}\) trapezoidal method. Apparent oral clearance of efavirenz (CL/F) was calculated by dividing the administered efavirenz daily dose (600 mg) by the AUC\(_{0-24\text{h}}\). Clearance was normalized to body weight (CL/F/W) measured at study entry in kilograms.
Enzymes and nuclear receptor genotyping

Subjects were genotyped for CYP2B6*6 (rs3745274), *16 (rs28399499), CYP2A6 *9B, (rs8192726), CYP2A6 *17 (rs28399454), pregnane X receptor (PXR, NR1I2) 63396C→T SNP (rs2472677) and constitutive androsane receptor (CAR, NR1I3) G→A SNP (rs2307424) using Applied Biosystems kits as we previously described. Genotypes for the (UGT2B7*2; rs7439366), UGT2B7*1c (rs28365062) and for PXR variants rs3732360C→T were determined by genomic PCR amplification and sequencing as previously described with minor modifications (9, 16, 31). The SNPs were selected because they have been previously shown to be associated with efavirenz plasma concentrations or effect (18, 19, 43), as well as CYP3A4 and CYP2B6 activity (12, 31). Composite CYP2B6 516/983-genotype was defined by the number of minor allele polymorphisms of CYP2B6 516G→T and 983C→T SNPs (0 = extensive metabolizer; 1 = intermediate metabolizer and 2 = slow metabolizer) (37).

Statistical analysis

Univariate analyses of association between patient factors and efavirenz AUC_{0-24} hour were assessed by t-test or Mann-Whitney rank sum test (sex, race, and history of alcohol use), analysis of variance ANOVA (three genotype groups) or Spearman rank order correlation test (age, body weight and body mass index). Arithmetic means of efavirenz pharmacokinetic parameters including T_{max}, C_{max}, C_{24h}, AUC_{0-24} hour, apparent oral clearance (CL/F), and weight-adjusted apparent oral clearance (CL/F/W) were compared by the use of paired t-test (Wilcoxon signed rank test, if data failed normality test). Geometric mean ratios (efavirenz plus rifampin/efavirenz alone) and 90% confidence intervals (90% CI) of log-transformed data were calculated using Win Nonlin Professional v 5.2.1 (Pharsight Corporation, Cary, NC). Finally, differences in demographic and genetic factors between patients with efavirenz C_{24h} concentration < 1000
ng/mL (considered sub-therapeutic) and > 1000 ng/mL where compared by t-test or Mann-Whitney rank sum test (continuous data) or Chi-square or Fischer Exact test (categorical data). A $P$ value < 0.05 was considered significant.

**RESULTS**

**Efavirenz pharmacokinetics in the study population**

Of the 13 volunteers who were initially enrolled, one subject developed a vasovagal episode during blood sampling and did not complete the study. Another subject (ID: B3) had detectable efavirenz concentrations when efavirenz was administered alone but undetectable concentrations with rifampin coadministration. Although medication dosing was observed on the day of PK sampling, we could not rule out medication non-adherence and the individual was excluded from further analysis.

The final study population of 11 subjects included 6 Caucasians and 5 African-Americans and 6 females and 5 males. The mean (SD) age was 42.6 (7.5) years, body weight was 76.9 (18.9) kilograms, and body mass index (body mass index) was 26.9 (5.7) kg/m$^2$. The coefficient of variation (CV) in efavirenz plasma AUC$_{0-24\,\text{h}}$ was 50% and 56% in the absence and presence of rifampin, respectively. The distribution of efavirenz plasma AUC$_{0-24\,\text{h}}$ in the absence and presence of rifampin, as well as the relationship with subject factors are shown in Figures 1A and 1B, respectively. *UGT2B7*1c carriers compared to noncarrier had higher efavirenz AUC$_{0-24\,\text{h}}$ in the basal state (mean, 110489 vs. 49496, $P < 0.001$), as well as in the induced state (mean, 93415 vs. 41773, $P = 0.006$). *CYP2B6* 516/983-genotype status was significantly associated with efavirenz AUC$_{0-24\,\text{h}}$ in the induced state (mean: 92507, 46200 and 29853 ng/mL for subjects with slow, intermediate and fast metabolizer genotypes, respectively; $P = 0.024$). A similar
relationship was observed in the basal state but the differences between genotype group did not reach statistical significance ($P = 0.057$). There was an inverse relationship between body weight and efavirenz AUC$_{0-24\text{h}}$ in the induced state (correlation coefficient = –0.764; $P = 0.005$). Age, body mass index, sex, ethnicity and other genetic factors evaluated were not significantly associated with efavirenz AUC$_{0-24\text{h}}$ in either treatment periods ($P > 0.05$) (Figure 1).

**Effect of rifampin on efavirenz pharmacokinetics**

The plasma concentration-time profile for efavirenz in the absence and presence of rifampin is shown in Figure 2. Mean efavirenz plasma concentrations at all sampling time points were lower when efavirenz was administered with rifampin but there was wide inter-subject variability. Efavirenz mean $C_{24\text{h}}$ and AUC$_{0-24\text{h}}$ values were lower with rifampin coadministration than when efavirenz was administered alone (1722 vs. 2116 ng/mL, $P = 0.014$) and (55810 vs. 66251 ng*h/mL, $P = 0.010$), respectively. The differences in mean values between the two periods for $T_{\text{max}}$, $C_{\text{max}}$ and CL/F were not significant (data not shown in tables).

Similar trends were observed in the pharmacokinetics parameters in the absence and presence of rifampin when we excluded the subject W6, in whom the C$_{24\text{h}}$ sample was lost during the period II (data not shown). The geometric means, and geometric mean ratios (on:off rifampin) with 90% CI for the efavirenz plasma pharmacokinetic parameters are summarized in Table 1. The geometric mean ratio (90%IC) for $C_{\text{max}}$ and AUC$_{0-24\text{h}}$ were 0.84(0.71, 1.00) and 0.82 (0.72, 0.92), respectively. The result did not change significantly after a repeat analysis with subject W6 excluded (data not shown).

Individual efavirenz AUC$_{0-24\text{h}}$ ratios (on: off rifampin) ranged from 0.55 to 1.18 (Figure 3), with an arithmetic mean (SD) of 0.84 (0.18). There was a trend towards inversely relationship between the AUC$_{0-24\text{h}}$ ratio and body weight (correlation coefficient = – 0.591, $P =$
0.051). We found no significant relationship between AUC\textsubscript{0-24h} ratio and age, sex, ethnicity, and the genetic factors evaluated.

The changes in efavirenz AUC\textsubscript{0-24h} or weight-normalized CL/F between the two periods appeared to be minimal in a majority of the subjects (Figure 4). Ten subjects showed a 6 – 44% decrease in efavirenz AUC\textsubscript{0-24h} with rifampin cotreatment. Two subjects (W1 and W4) had slight increases in efavirenz AUC\textsubscript{0-24h} of 3 and 18%, respectively (Figure 4A). The percent change in CL/F from baseline varied from +1041% to –24% [mean (SD) of 139.1% (313.8%)]. Three subjects (W001, W004, and B006) had paradoxical decreases in efavirenz CL/F of 24.1, 21.1 and 14.0% with rifampin cotreatment compared to when efavirenz was administered alone (Figure 4B).

Factors associated with efavirenz C\textsubscript{24h} < 1000 ng/mL in the presence of rifampin

Five of the 11 subjects had efavirenz C\textsubscript{24h} < 1000 ng/mL during rifampin cotreatment, while only one subject had efavirenz C\textsubscript{24h} < 1000 ng/mL in the absence of rifampin. Given that the suggested minimum effective plasma concentration of efavirenz is 1000 ng/mL (28, 32), we sought to identify individual factors associated with efavirenz C\textsubscript{24h} < 1000 ng/mL during rifampin coadministration. Body weight-adjusted efavirenz dose in milligram/kilogram, and lower baseline efavirenz plasma AUC or C\textsubscript{24h} were significantly associated with efavirenz C\textsubscript{24h} < 1000 ng/mL (Table 2). Both subjects with CYP2B6 516/983 fast metabolizer genotypes in the study population had C\textsubscript{24h} < 1000 ng/mL and two of the three individuals with slow metabolizer genotypes had C\textsubscript{24h} > 4000 ng/mL, which is considered the upper limit of the therapeutic range (Table 2).
Safety and tolerability

The administration of efavirenz alone or with rifampin for 8 days was well tolerated in this study. Central nervous system (CNS) side effects of efavirenz were evaluated using the CNS symptoms experience questionnaire designed for the AIDS Clinical Trials Group study A5097s (7). The self-administered questionnaire included 34 questions (scaled from 0 – 4) with range of scores from 0 – 136. The change in CNS symptoms test score when efavirenz was administered alone varied from – 2 to + 16 with four subjects showing no changes in scores. During the period of efavirenz and rifampin coadministration the changes in test scores varied from – 16 to + 38, with two subjects reporting no changes. One subject (ID B004) with the highest efavirenz exposure had the highest increase in symptoms test scores during both periods. Symptoms that were considered by Subject B004 to be extreme (scale 4) included restless sleep, waking up a lot, intense dreams and trouble going to sleep. There was no discontinuation of study medications due to adverse events by any of the study subjects.

DISCUSSION

The findings of this study indicate a modest decrease in mean efavirenz plasma exposure with rifampin coadministration in the subjects as a whole, as well as a wide variability in response. The mean decrease in efavirenz AUC\(_{0-24h}\) of 16% in this crossover study is numerically lower than the 26% reduction reported in a healthy volunteer study (2) and the 24% decrease in Spanish HIV/TB co-infected patients (24). However, the mean decrease in efavirenz AUC\(_{0-24h}\) was significant (\(P < 0.05\)) in our and the previous healthy volunteer study (2) but was not in the study among HIV/TB co-infected patients (24). A common finding in the previous pharmacokinetic studies (2, 24), and ours is wide inter-individual variability in the rifampin
effect, with some subjects showing no decrease in efavirenz plasma exposure with rifampin therapy. In our study, the reduction in efavirenz AUC$_{0-24h}$ with rifampin ranged from a decrease of 44% to an increase of 18%. The large deviation (both positive and negative) from the average in the current and previous studies might explain the modest effect of rifampin in the study populations as a whole. The apparent lack of induction of efavirenz metabolism by rifampin-containing anti-TB therapy in some subjects is contrary to expected effect of rifampin but it is consistent with the bimodal effects of rifampin-containing anti-TB therapy on efavirenz plasma concentrations observed in HIV/TB co-infected patients (13, 17, 35).

The clinical implications of the induction or lack of induction effect of rifampin on efavirenz pharmacokinetics are not entirely clear. Many patients achieve adequate efficacy and tolerability with efavirenz 600 mg or 800 mg daily during coadministration with rifampin (3, 23, 27). However, efavirenz 800 mg daily was associated with a high frequency of central nervous system (CNS) and hepatic toxicities associated with supra-therapeutic plasma concentrations in black native Africans (3). Furthermore, a reduced efavirenz dose to 200 mg daily or discontinuation of efavirenz has been necessary in some HIV/TB co-infected patients during concurrent rifampin-containing TB therapy because of intolerable CNS toxicities (14, 15, 41). In contrast, other HIV/TB co-infected patients have required increased doses up to 1600 mg daily to achieve desired plasma concentrations and virologic suppression during concurrent therapy (4). The above-mentioned clinical studies or case reports indicate that a better understanding of the individual differences in the efavirenz pharmacokinetic response to rifampin coadministration is necessary for rational decisions about efavirenz dose-adjustment with rifampin therapy, as one dose adjustment will not fit all patients.
Nearly one-half of the subjects in this study had efavirenz trough concentrations considered to be sub-therapeutic during the period of rifampin coadministration but only one of the 11 patients had sub-therapeutic trough concentration in the absence of rifampin. Although the minimum effective efavirenz plasma concentration is still controversial, our findings suggest that some but not all individuals are likely under-dosed using the current standard fixed dose. Both $CYP2B6$ 516/983 “fast metabolizers” as well as higher body weight appeared to predict a lower efavirenz concentration with rifampin therapy. The data from this controlled pharmacokinetic study support the findings of several studies demonstrated a relationship between efavirenz concentrations and body weight (24, 26, 39), as well as with $CYP2B6$ 516G→T polymorphisms (8, 19, 20, 34, 40).

The main limitation of our study was the small number of subjects with known functional genetic variants. This limited our ability to draw any firm conclusions about lack of association with some of the genetic factors especially for PXR and CAR. We also did not assay for efavirenz metabolites and could not use the metabolite-to-parent AUC ratio as an index of the activity of enzymes mediating the main or accessory pathways in each subject to assess the magnitude of induction. Notwithstanding, this controlled crossover study provides insights into the inter-individual variability in response to the induction effects of rifampin on efavirenz disposition. Individuals with larger body weight, $UGT2B7*1c$ noncarriers and those with $CYP2B6$ extensive metabolizing genotype had lower efavirenz $AUC_{0-24h}$ with rifampin coadministration. Our findings suggest that genetic factors and body weight may be important factors and should be taken into consideration for developing dosing algorithms or when designing larger studies to determine the appropriate dose of efavirenz with rifampin coadministration.
Acknowledgements

This research was supported in part by a K23 developmental award (NIH K23 AI071760) to Dr Kwara and by the AIDS Clinical Trials Group funded by the National Institute of Allergy and Infectious Diseases (U01 AI069472). Dr. Court was supported by NIH grant R01-GM-61834 from the National Institute of General Medical Sciences (Bethesda, MD).

We thank the study participants, as well as the nurses and staff of at the ACTG Unit and the Immunology Center Laboratory for all their valuable assistance in recruitment, evaluation of subjects as well as obtaining, handing and processing of the pharmacokinetic samples. We also thank Dr. Heyward Hull at UNC for guidance on the statistical analysis. The University of North Carolina at Chapel Hill, Center for AIDS Research #9P30 AI50410, Clinical Pharmacology and Analytical Chemistry Core analyzed the efavirenz concentrations. The content is solely the responsibility of the authors and does not necessarily represent the official views of the funding organizations.
Conflict of Interest:

AK and ADMK previously received research grants from Bristol-Myers Squib not related to this research. KTT receives research funding from Merck and Co., Inc and GlaxoSmithKline. JBD, PP, JK, MHC, and DJG report no conflicts of interest.
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Table 1. Geometric mean, geometric mean ratio (90% confidence interval) of efavirenz steady-state pharmacokinetic parameter estimates in the absence and presence of efavirenz in healthy volunteers

<table>
<thead>
<tr>
<th>Pharmacokinetic parameter</th>
<th>Geometric mean</th>
<th>Geometric mean ratio (EFV+RIF/EFV)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>EFV alone (N = 11)</td>
<td>EFV/RIF (N = 11)</td>
</tr>
<tr>
<td>T&lt;sub&gt;max&lt;/sub&gt; (h)</td>
<td>2.46 (2.10, 2.89)</td>
<td>2.30 (1.56, 3.39)</td>
</tr>
<tr>
<td>C&lt;sub&gt;max&lt;/sub&gt; (ng/ml)</td>
<td>4571 (652, 5721)</td>
<td>3882 (3034, 4966)</td>
</tr>
<tr>
<td>C&lt;sub&gt;24h&lt;/sub&gt; (ng/ml)</td>
<td>1766 (1262, 2472)</td>
<td>1435 (1026, 2009)</td>
</tr>
<tr>
<td>AUC&lt;sub&gt;0-24h&lt;/sub&gt; (ng*hr/ml)</td>
<td>59429 (45604, 77466)</td>
<td>48865 (36475, 65464)</td>
</tr>
<tr>
<td>CL/F (mL/hr)</td>
<td>2958 (1380, 6339)</td>
<td>4656 (3112, 6966)</td>
</tr>
</tbody>
</table>

CI, confidence interval; EFV, efavirenz; RIF, rifampin; N, number of subjects; C<sub>max</sub>, peak concentration; T<sub>max</sub>, time to C<sub>max</sub>; C<sub>24h</sub>, 24 hour post-dose concentration; AUC<sub>0-24h</sub>, total area under the curve from time 0-24 hours; CL/F, apparent oral clearance.
Table 2. Comparison of characteristic of patients who had 24-hour post-dose efavirenz concentration < versus > 1000 ng/mL during concurrent administration with rifampin

<table>
<thead>
<tr>
<th>Characteristic</th>
<th>$C_{24h} &lt; 1000$</th>
<th>$C_{24h} &gt; 1000$</th>
<th>$P$</th>
</tr>
</thead>
<tbody>
<tr>
<td>Mean (SD) age (years)</td>
<td>42.2 (5.5)</td>
<td>43.0 (9.4)</td>
<td>0.871</td>
</tr>
<tr>
<td>Mean (SD) body weight (kg)</td>
<td>88.6 (19.0)</td>
<td>67.2 (13.3)</td>
<td>0.052</td>
</tr>
<tr>
<td>Mean (SD) body mass index (kg/m2)</td>
<td>29.0 (5.6)</td>
<td>25.2 (5.8)</td>
<td>0.294</td>
</tr>
<tr>
<td>Dose/body weight (mg/kg)</td>
<td>7.0 (1.2)</td>
<td>9.2 (1.6)</td>
<td>0.034</td>
</tr>
<tr>
<td>Sex</td>
<td></td>
<td></td>
<td>0.080</td>
</tr>
<tr>
<td>Male</td>
<td>4 (80.0%)</td>
<td>1 (20.0%)</td>
<td></td>
</tr>
<tr>
<td>Female</td>
<td>1 (16.7%)</td>
<td>5 (83.3%)</td>
<td></td>
</tr>
<tr>
<td>Race</td>
<td></td>
<td></td>
<td>0.567</td>
</tr>
<tr>
<td>African-American</td>
<td>3 (60.0%)</td>
<td>2 (40.0%)</td>
<td></td>
</tr>
<tr>
<td>Caucasian</td>
<td>2 (33.3%)</td>
<td>4 (66.7%)</td>
<td></td>
</tr>
<tr>
<td>CYP2B6 516/983 genotype</td>
<td></td>
<td></td>
<td>0.084</td>
</tr>
<tr>
<td>Extensive</td>
<td>2 (100.0%)</td>
<td>0 (0.0%)</td>
<td></td>
</tr>
<tr>
<td>Intermediate</td>
<td>3 (50.0%)</td>
<td>3 (50.0%)</td>
<td></td>
</tr>
<tr>
<td>Slow</td>
<td>0 (0.0%)</td>
<td>3 (100%)</td>
<td></td>
</tr>
<tr>
<td>History of alcohol use</td>
<td></td>
<td></td>
<td>1.00</td>
</tr>
<tr>
<td>No</td>
<td>2 (50.0%)</td>
<td>2 (50.0%)</td>
<td></td>
</tr>
<tr>
<td>Yes</td>
<td>3 (42.9%)</td>
<td>4 (57.1%)</td>
<td></td>
</tr>
</tbody>
</table>
Mean (SD) baseline efavirenz AUC (ng*h/mL) | 44965 (14720) | 83768 (34655) | 0.046
Mean (SD) baseline efavirenz C\textsubscript{24h} (ng/mL) | 1163 (336) | 2910 (1555) | 0.037
Mean (SD) AUC\textsubscript{0-24h} ratio (on/off rifampin) | 0.75 (0.16) | 0.92 (0.164) | 0.104

SD, standard deviation, C\textsubscript{24h}, 24-hour post-dose concentration; AUC\textsubscript{0-24h}, total area under the curve from time 0 to 24 hours.
Figure legends

Figure 1. Distribution of efavirenz AUC$_{0-24h}$ in increasing order and relationship with individual factors in the absence (panel A) and presence (panel B) of rifampin. Composite $CYP2B6$ 516/983-genotype was defined by the number of minor allele polymorphisms of $CYP2B6$ 516G→T and 983C→T SNPs. Relationship between subject factor and efavirenz AUC$_{0-24h}$ is significant (P < 0.05). Differences between groups were examined by Mann-Whitney rank sum test (2 genotype groups) or by ANOVA (3 genotype groups) and relationship with body weight was examined by Spearman rank order correlation test. Shading code for genetic factors: clear, homozygous reference; gray, heterozygous; black is homozygous variant or carrier.

Figure 2. Mean efavirenz concentration-time profile in 11 healthy volunteers measured on day-8 of the treatment period either in the absence (black circles) or presence (clear circles) of rifampin. Error bars indicate standard deviation of the mean.

Figure 3. Distribution of efavirenz AUC$_{0-24h}$ (with rifampin/no rifampin) ratio in increasing order and relationship with individual factors. Composite $CYP2B6$ 516/983-genotype was defined by the number of minor allele polymorphisms of $CYP2B6$ 516G→T and 983C→T SNPs. Shading code for genetic factors: clear, homozygous reference; gray, heterozygous; black is homozygous variant or carrier.

Figure 4. Efavirenz AUC$_{0-24h}$ (panel A), and weight-adjusted apparent oral clearance (panel B) in 12 healthy volunteers in the absence and presence of rifampin. Efavirenz and rifampin were administered at standard dosage and patients were in steady state. Rifampin co-administration
caused a mean reduction in AUC$_{0-24h}$ by 16% and a mean increase in apparent oral clearance by 139%. Open circles with dotted line represents individuals and triangle with solid line is the median.
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