**Evaluation of universal versus genotype-guided efavirenz dose reduction in pregnant women using population pharmacokinetic modeling**

Adeniyi OLAGUNJU1,2\*#, Alessandro SCHIPANI2#, Oluseye BOLAJI1, Saye KHOO2, Andrew OWEN2

1Faculty of Pharmacy, Obafemi Awolowo University, Ile-Ife, Nigeria

2Department of Molecular and Clinical Pharmacology, University of Liverpool, 70 Pembroke Place, Liverpool, L69 3GF, United Kingdom

#Adeniyi Olagunju and Alessandro Schipani contributed equally to this manuscript.

**Running title:** PopPK of 400 mg efavirenz in pregnancy

**\*Correspondence:** Adeniyi Olagunju, PhD, Faculty of Pharmacy, Obafemi Awolowo University, Ile-Ife, Nigeria. (Tel: +2348026640439. email: aeolagunju@oauife.edu.ng).

**Objectives:** Lack of data on the pharmacokinetics of efavirenz in pregnant women at the 400 mg reduced dose currently prevents universal roll out. Population pharmacokinetic modeling was used to explore pharmacokinetic end-points at 200, 400, and 600 mg daily doses in pregnant women stratified by *CYP2B6* metabolic status.

**Methods:** The analysis was based on 252 plasma efavirenz concentrations from 77 pregnant women (77 sparse, 175 intensive) who received antiretroviral regimens containing 600 mg efavirenz. The model was developed using NONMEM®. The effect of genetics was investigated and concentration-time courses at steady state were simulated for individuals (n = 1000 each) classified as *CYP2B6* slow, intermediate, and fast metabolisers at 200, 400, and 600 mg daily doses.

**Results:** At a 400 mg reduced dose, predicted mean (90% CI) mid-dose efavirenz concentration (C12) was 2.24 µg/mL (0.89, 4.18) in pregnant women classified as slow metabolisers, compared with 0.87 µg/mL (0.34, 1.64) in intermediate and 0.78 µg/mL (0.30, 1.47) in fast metabolisers. C12 was below the 0.47 µg/mL threshold determined within the ENCORE 1 trial in 10% at 400 mg, 4.6% at 600 mg, and 3.4% with genotype-guided dosing. The 4.0 µg/mL toxicity threshold was exceeded in 4.5% at 400 mg, 13.5% at 600 mg, and 5.2% with genotype-guided dosing.

**Conclusions:** These data provide context for the ongoing debate about reduction in efavirenz dose to 400mg during pregnancy, and should be interpreted alongside the lower toxicity expected with the lower dose. Additional research is required to investigate genotype-guided dose reduction in pregnant women.

The current WHO guidelines on the use of antiretroviral drugs for treating and preventing HIV infection recommend lifelong antiretroviral therapy (ART) in all adults living with HIV, including pregnant women, regardless of CD4 cell count or clinical stage of disease.1 The benefits associated with universal lifelong ART for prevention of mother-to-child transmission (PMTCT) of HIV are believed to outweigh the risks.2 Like the Option B and B+ from which it developed, this approach is expected to further reduce the risk of MTCT, improve maternal health benefits, and reduce the risk of HIV transmission to sexual partners. However, implementation of universal ART coverage, though urgent, will significantly increase the funding required for diagnosis, drugs, and personnel. This has generated interest in strategies for reducing the cost of antiretroviral drugs, including optimisation of the manufacturing process, innovative formulations, and dose reduction.3

For instance, reduced doses of indinavir/ritonavir (400/100 mg versus 800/100 mg twice daily) and atazanavir/ritonavir (200/100 versus 300/100 mg once daily) were shown to have the same efficacy and lower side effects compared with the standard doses.4, 5 Reduced doses of efavirenz (200 mg and/or 400 mg) were similarly reported to result in sustained virological suppression and reduced side effects despite lower plasma concentrations compared with the standard 600 mg dose.6, 7 The non-inferiority of the 400 mg daily reduced dose compared with the standard dose was confirmed in the ENCORE1 study,8 resulting in its adoption by the WHO as part of alternative first-line ART in the 2016 guideline. However, pregnant women, patients with HIV/TB co-infection and adolescents younger than 12 years old are still excluded from this recommendation because of lack of efficacy and safety data.1 Efavirenz-based regimens are preferred as first-line ART across different populations, including pregnant women where efavirenz safety has been demonstrated.9

Pregnancy is known to alter the pharmacokinetics of many drugs, including certain antiretroviral (ARV) drugs.10 In a recent pharmacokinetic study, we reported significant differences in the magnitude of pregnancy-induced changes in efavirenz pharmacokinetics among HIV positive women stratified by a single nucleotide polymorphism (SNP; 516G>T; rs3745274) in efavirenz disposition gene, *CYP2B6*.11 Pregnancy was associated with an approximately 100% increase in efavirenz apparent clearance (Cl/F) in women with the *CYP2B6* 516GG genotype compared with postpartum control, resulting in median (range) minimum plasma concentration (Cmin) of 0.59 µg/ml (0.43-0.92), compared with the recommended 1.0 µg/mL.11 However, the 1.0 µg/mL target was determined in patients with older regimens almost two decades ago,12 and its validity is dubious given the lack of evidence of therapeutic failure in patients with plasma concentrations below 1.0 µg/mL in a number of studies.13, 14 More recently, the non-inferiority of 400 mg dose of efavirenz compared to the usual 600 mg dose was demonstrated in the ENCORE 1 trial,15 despite plasma concentrations below 1.0 µg/mL in some patients.16 Consequently, reduction of efavirenz dose to 400 mg has been suggested as a way of saving cost while maintaining efficacy and reducing adverse events.3

Although the 400 mg reduced dose of efavirenz has not been evaluated in pregnant women, observation of significantly lower plasma concentrations in pregnant women with the *CYP2B6* 516GG compared with other genotypes11, 17 suggest the need for a cautious evaluation of this dose before roll out in this population.18 An ongoing study (ClinicalTrials.gov ID: NCT02499874) will evaluate key pharmacokinetic parameters of the 400 mg reduced dose of efavirenz in a cohort of pregnant women on suppressive antiretroviral therapy (n = 25; plasma HIV-1 RNA < 50 copies/mL). However, the study is not adequately powered to explore differences between genotype groups, and complementary approaches are therefore needed to guide prospective efficacy trials towards the adoption of the 400 mg reduced efavirenz dose during pregnancy.

In the present study, we used a population pharmacokinetic modelling approach to compare plasma exposure resulting from a universal roll out of 400 mg reduced dose of efavirenz, and *CYP2B6* genotype-guided dose reduction in pregnant women. The analysis is based on data from a previously published study, Clinicaltrials.gov ID: NCT02269462.11

**METHODS**

**Clinical Study Population and Design**

HIV positive pregnant women were recruited from three hospitals in Benue State, Nigeria: Bishop Murray Medical Centre, Makurdi; St Monica’s Hospital, Adikpo; and St Mary’s Hospital, Okpoga. Details about study protocol have been previously reported11 and available at Clinicaltrials.gov (ID: NCT02269462). In brief, the clinical study was conducted in two phases in HIV positive pregnant women receiving efavirenz in combination with two nucleoside reverse transcriptase inhibitors. Those taking other drugs or herbs with known or uncertain interaction with antiretrovirals (e.g. anti-tuberculosis drugs) were excluded. The purpose of the preliminary phase was to identify SNPs associated with efavirenz concentrations during pregnancy. In the second phase, the SNP independently associated with efavirenz plasma concentrations with the highest predictive power (*CYP2B6* 516G>T; rs3745274) was used to stratify patients into groups and randomly selected patients from each group were invited for the intensive pharmacokinetic phase.

**Sample Collection, SNP Genotyping and Efavirenz Quantification**

As previously reported,11 samples were collected at a single time point after dose in the preliminary phase for DNA extraction and efavirenz quantification. Genotyping was conducted for *CYP2B6* 516G>T (rs3745274), *CYP2B6 983T>C* (rs28399499) and seven other SNPs using TaqMan® assays (Life Technologies Ltd, Paisley, Renfrewshire, UK). The influence of the genotype, was tested on efavirenz CL by dividing the patient’s population into 3 sub-groups based on combined effect of *CYP2B6* 516G>T and 983T>C. Individuals with two or more variant alleles (516GT plus 983TC, 516TT plus 983TT, or 516TT plus 983TC) were classified as ‘slow’ metabolisers, those with one variant allele (516GT plus 983TT, or 516GG plus 983TC) were classified as ‘intermediate’ metabolisers, and those with no variant allele (516GG plus 983TT) were classified as ‘fast metabolisers’. The current data set had only wild type and heterozygous individuals for the rare *CYP2B6* 983T>C. Intensive pharmacokinetic samples (7 per subject) were collected between 0.5 and 24 hours after an observed evening dose of 600 mg efavirenz. Efavirenz was quantified using a validated liquid chromatography-mass spectrometry method.19

**Population Pharmacokinetic-Pharmacogenetic Model Development**

The model was developed using NONMEM® (ICON, version VII 2.0). The model building strategy was as follows: one- and two- compartment models with first- or zero-order absorption without and with lag-time were fitted to the data using the first order conditional method of estimation. Proportional, additional, and combined proportional and additional error models were evaluated to describe residual variability. Inter-occasion variability was also tested. The minimal objective function value (OFV; equal to -2 log likelihood) was used as a goodness-of-fit metric with a decrease of 3.84 corresponding to a statistically significant difference between models (*P* = 0.05, χ2 distribution, one degree of freedom). Residual plots were also examined. Exponential errors following a log-normal distribution were assumed for the description of inter-individual variability in pharmacokinetic parameters, as shown in equation 1.

*θ*i = θ1 \*exp (ηi) …………………………………………………………………………………………………………………. (1)

where θi is the pharmacokinetic parameter of the ith individual; θ1 is the population parameter estimate; and ηi is the inter-individual variability with a mean of zero and variance ω2.

Once the appropriate structural model was established, the following covariates were explored: age, body weight, gestational age, and SNPs. An allometric weight model was applied to standardize the pharmacokinetic parameters using a standard weight (WTstd) of 70 kg. An allometric weight model for clearance parameters is given by CLwt=(WT/WTstd)0.75 and for volume parameters is given Vwt=(WT/WTstd)1 where CLwt and Vwt are the weight functions for clearance parameters and volume of distribution (V) parameters, respectively, and WT is the individual weight value.20, 21 Continuous variables were modelled using a power model with normalized covariate:

 *θxi* = (*θx*×(COVi/COVmedian) *θ*cov)\*exp (*ηxi*) ……………………………………………………………………………. (2)

where *θxi*is pharmacokinetic parameter “x” in the *i*th individual and *θ*x is the population parameter estimate as previously. In equation 2 (for continuous covariates) COVi is thevalue of the covariate for the *i*th individual, COVmedian is the median value in the population dataset, *θ*cov is theexponent describing the covariate effect. Graphical methods were used to explore the relationship of covariates with individual predicted pharmacokinetic parameters. Each covariate was introduced separately into the model and only retained if inclusion in the model produced a statistically significant decrease in OFV of 3.84 (*P* ≤ 0.05). A backwards elimination step was then carried out once all relevant covariates were incorporated and covariates were retained if their removal from the model produced a significant increase in OFV (> 6.63 points; *P* ≤ 0.01, χ2 distribution, one degree of freedom).

To perform a visual predictive check, 1000 datasets were simulated using the parameter estimates defined by the final model with the SIMULATION SUBPROBLEMS option of NONMEM®. Datasets were simulated for efavirenz 600 mg daily standard dose. From the simulated data, 90% prediction intervals (P5–P95) were constructed and superimposed on observed data from the original dataset. At least 90% of data points within the prediction interval (5% above and below) was considered an adequate model. In addition, in order to confirm the stability and robustness of the model, a bootstrap re-sampling was used. Bootstrapping was performed with the software package Perl-speaks-NONMEM.22 The median values and 95% confidence intervals for the parameter estimates were obtained from 200 bootstrap replicates of the original data set and compared with the original population parameters.

To investigate the different dosing regimen scenarios (400 mg daily and 200 mg daily), pharmacokinetic simulations were performed and 90% prediction intervals of the simulated efavirenz concentrations for each category were plotted. For these simulations, dose-linear pharmacokinetics was assumed as has previously been demonstrated across human doses of this drug.23

**RESULTS**

**Data Set**

The analysis was based on 252 plasma efavirenz concentrations from HIV positive pregnant women receiving regimens containing 600 mg efavirenz daily for ≥ 4 weeks. Of these, 77 were sparse pharmacokinetic samples from 77 women, and 175 were intensive pharmacokinetic samples from 25 women stratified based on their genotypes. Genotyping was successful for all 77 subjects. Patients’ demographics and genotype frequencies are shown in **Table 1.** This cohort and the data set on which the present analysis is based were previously described.11 Concentration-time profiles for individual patients in the intensive pharmacokinetic phase are shown in **Figure 1**; C12 was below 1.0 µg/mL in 20% (5/20) of patients and Cmin in 44% (11/25). The median (range) efavirenz AUC0-24, Cmax, C12, and Cmin were 42.6 µg.h/mL (21.7-203), 3.5 µg/mL (1.3-14.4), 1.6 µg/mL (0.78-8.6), and 1.0 µg/mL (0.43-5.2), respectively.

**Population Pharmacokinetic Analysis**

Efavirenz pharmacokinetic was best described by a one-compartment model with first-order absorption and first-order elimination. A one-compartment model with zero-order absorption or a two-compartment model did not improve the fit. In the model, residual variability was best described by a proportional structure; the inclusion of an additive structure did not improve the model. Inter-individual random effects were described by an exponential model which was supported for CL/F, apparent volume of distribution (V/F) and absorption constant (ka). In the basic model the mean population estimates for CL/F, V/F and absorption constant were 12.7 L/h, 268 L and 0.58 h-1, respectively; the inter-individual variability in CL/F, V/F and absorption rate expressed by the coefficient of variation (CV%) were 63.3, 23.5 and 88.2 respectively. The residual variability was 0.08.

A total of 5 covariates (age, body weight, gestational age, *CYP2B6* 516G>T and *CYP2B6* 983T>C) were analysed using a stepwise backward elimination. The final covariate model is detailed in **Table 2**. Only the genetic covariates were statistically significant. The impact of *CYP2B6* genotypes on CL/F was used to categorise the population into three sub-groups based on combined effect of 516G>T and 983T>C (Table 2). The inclusion of genotypes in the final model decreased the OFV by 57 (P < 0.001, degree of freedom = 5). A summary of the final population estimates is presented in **Table 2**. The inclusion of the genotypes decreased the CL/F inter-individual variability by 22.2%. A 90% prediction interval was generated from 1000 simulations for efavirenz 600 mg once daily, with the covariate values of those individuals used in the building process (**Figure 2**). Visual predictive checks for the final model showed that predicted and observed data were in adequate agreement.

**Dose Optimisation Simulations**

To investigate the effect of genetics on efavirenz concentration at the end of the dosing interval (Ctrough), we simulated pharmacokinetic data for efavirenz, stratified for *CYP2B6* 516G>T and 983T>C genotype. Simulated concentration-time courses at steady state were generated for the 200 mg, 600 mg and 400 mg daily doses. The simulations were carried out with a population of individuals classified as slow, intermediate, and fast metabolisers (**Figure 3**). For the 400 mg once daily reduced dose simulations, mean (90% CI) C12 was 2.24 µg/mL (0.89, 4.18) in pregnant women classified as slow metabolisers, compared with 0.87 µg/mL (0.34, 1.64) in intermediate metabolisers and 0.78 µg/mL (0.30, 1.47) in fast metabolisers. For the standard dose of 600 mg once daily, C12 was 3.37 µg/mL (1.35, 6.31) in slow, 1.31 µg/mL (0.51, 2.48) in intermediate, and 1.17 µg/mL (0.45, 2.21) in fast metabolisers (**Table 3**). Decreasing the daily dose to 200 mg resulted in C12 of 1.14 µg/mL (0.45, 2.10) in slow, 0.44 µg/mL (0.17, 0.82) in intermediate, and 0.39 µg/mL (0.15, 0.95) in fast metabolisers.

At the 400 mg reduced dose, approximately 45% of women were predicted to have C12 below the questionable 1.0 µg/mL cut-off compared with 23% at the standard 600 mg reduced dose. About 10% were predicted to have C12 below 0.47 µg/mL (recommendation based on data from the ENCORE1 trial24) at the 400 mg dose compared with only 3% at 600 mg, increasing to 42% when the dose was further reduced to 200 mg (**Table 3**). Also, the proportion of patients with predicted C12 above 4.0 µg/mL decreased from 13.5% at 600 mg to 4.6% at the 400 mg reduced dose. Slow metabolisers were the most at risk with both doses (**Table 3**).

**DISCUSSION**

One of the key issues delaying the universal roll out of the 400 mg reduced dose of efavirenz is the lack of data on its pharmacokinetics, efficacy and safety in pregnant women.1, 18 The analysis presented here has provided an early insight into efavirenz exposure in the context of the 400 mg reduced dose in this population. Universal adoption of the 400 mg reduced dose was predicted to achieve mean (90% CI) C12 of 0.87 µg/mL (0.34, 1.64) and 0.78 µg/mL (0.30, 1.47) in pregnant women classified as intermediate and fast metabolisers, respectively, compared with median (range) of 2.14 µg/mL (0.579-7.89) and 1.60 µg/mL (0.734-3.65) reported in non-pregnant adults.24 This is below the 1.0 µg/mL threshold previously associated with virological suppression.12 However, no association was found between pharmacokinetic parameters and achieving viral load below 200 copies/mL at 96 weeks in the ENCORE1 trial. In a comprehensive analysis, the sensitivity/specificity of using C12 of 1.0 µg/mL for achieving plasma viral load below 200 copies/mL at 96 weeks was 97.1%/84. %, with a likelihood ratio of 6.24 Using a range of potential C12 cut-offs between 0.47 and 0.76 µg/mL also provided adequate sensitivity/specificity of 90% (100%/92.3% to 98.9%/ 92.3%) with a likelihood ratio of 13. The lower limits of the predictive interval for fast and intermediate metabolisers in the present study fall outside this range. In fact, a comparison of the AUC0-24 and C12 values across doses and metabolising groups between the ENCORE 1 trial (online resource 2) and the present analysis indicates about 50% reduction in pregnant women.24

However, about 10% of pregnant women (16, 13 and 1.2% of fast, intermediate and slow metabolisers, respectively) were predicted to achieve plasma concentrations below 0.47 µg/mL at 400 mg, compared with 3% at the 600 mg dose (**Table 3**). Interestingly, data from the ENCORE1 trial showed no difference in time to virological suppression between patients in the 400 mg and 600 mg groups. Plasma HIV RNA was above 200 copies/mL at week 24 in about 10% (33/321) of patients in the 400 mg group and in 9.7% (30/309) in the 600 mg group, further reducing to 1.2% (4/321) and 2.9% (9/309), respectively, at week 36 with a mean difference of -0·02 log10 copies/mL (95% CI -0·14 to 0·10; p = 0·74.8 Therefore, the 0.47 µg/mL C12 cut-off appears reasonable for women who start ART during pregnancy for viral load below 200 copies/mL to be achieved at delivery. In fact, despite risks of Ctrough below 1.0 µg/mL in pregnant women with *CYP2B6* fast metaboliser status who received the standard 600 mg in two separate studies,11, 17 further analysis showed that median C12 was 1.01 µg/mL (range: 0.78-1.26) in one study 11, and no increased risk of MTCT was observed in the other.17

The risk of C12 below 0.47 µg/mL predicted for 10% of pregnant women who received the 400 mg reduced dose raises concerns, especially when therapy is started late in pregnancy. Further research is required to validate these predictions, but a dose reduction below 400 mg is unlikely to provide adequate exposure in pregnant women. This is important because suboptimal virological suppression in HIV positive pregnant women results in detectable viral load at delivery and increases the risk of MTCT.25, 26 For instance, viral load above 1000 copies/mL near delivery was associated with a 12-fold increased risk of MTCT in a European Collaborative Study.27 In a large multicentre US cohort study, Katz *et al.* reported detectable viral load at delivery in 13% of 671 women who initiated ART during pregnancy, increasing to 23.9% when started at third trimester, and identified gestational age at ART initiation, poor medication adherence, and treatment interruptions as factors increasing risk.28 Among South African women who started ART before pregnancy (n = 574), 13% reportedly had viral load above 1000 copies/mL.29 MTCT risks of 0.25, 2.0 and 8.5% were recently reported among women with viral load < 50, 50-1000 and > 1000 copies/mL, respectively, at delivery.30 Therefore, lack of virological suppression at delivery appears to be a major contributor to residual MTCT in women receiving ART. While it is still difficult to establish a pharmacokinetic target for PMTCT, the importance of undetectable viral load at delivery for PMTCT has been demonstrated in these studies. This highlights the need for continued caution with universal roll out of the 400 mg reduced dose of efavirenz in pregnant women.

Therefore, the present analysis indicates that a confirmation of adequate virological suppression in pregnant women classified as *CYP2B6* fast and intermediate metabolisers will be required before universal implementation of the 400 mg reduced dose. Meanwhile, where genotyping capacity already exists, retention of the 600 mg dose could be considered for fast and intermediate metabolisers to achieve C12 of 1.31 µg/mL (0.51, 2.48) and 1.17 µg/mL (0.45, 2.21), respectively, while the 400 mg dose will achieve C12 of 2.24 µg/mL (0.89, 4.18) in slow metabolisers during pregnancy. This approach will also reduce the proportion of patients (mainly slow metabolisers) at risk of early treatment discontinuation caused by CNS side effects due to plasma concentrations above 4.0 µg/mL; 5.2% with genotype-guided dosing compared to 13.5% with universal 600 mg dose, and 4.5% with universal 400 mg dose.31-33 In addition, only 3.4% of simulated pregnant women had C12 below 0.47 µg/mL with genotype-guided dosing, compared with 10% with universal 400 mg dose. Despite the difference in percentage of patients predicted to remain above this efficacy threshold, any future decision about implementation of the 400mg dose needs to consider that a) not all patients below this threshold will fail therapy, and b) these simulations do not take into account the incidence of CNS toxicity for either dose. It should be noted that CNS toxicity is more common in patients receiving 600mg,24 and this may subsequently impact adherence and therefore efficacy.34

Schackman *et al.* recently conducted cost-effectiveness analysis comparing the 600 mg standard dose with universal dose reduction to 400 mg, and genotype-guided dosing.35 Assuming equal efficacy, current standard dose increases lifetime cost by $18,500 and genotype-guided dosing was found to be more cost–effective. However, universal dose reduction to 400 mg was more cost-effective than both strategies under the most plausible scenarios. These findings are consistent with the findings of an earlier observational study.36 Similar cost-effectiveness analysis is now warranted to compare the current standard care with targeted genotype guided dose reduction and universal dose reduction to 400 mg in pregnant women. Such analysis should incorporate likely changes in rates of virological suppression with universal dose reduction in fast and intermediate metabolisers, as well as potential changes in available treatment options for pregnant women. Also, a value of information analysis to quantify the value of undertaking additional research to investigate the effectiveness of a genotype-guided dose reduction will help in prioritising research efforts.37

Limitations in the present study include lack of data on virological suppression which precludes pharmacokinetic-pharmacodynamic analysis, the limited number of patients in the different subgroups following stratification, and lack of information on patient adherence for the sparse pharmacokinetic data. Also, the absence of the *CYP2B6* 983CC genotype in the cohort means ultra-slow metabolisers are not represented. In addition, differences in MTCT risks between slow, intermediate and fast metabolisers could not be assessed because data on MTCT was not available from the original study. The narrow range of body weight (48-83 kg) should be taken into account in interpreting the absence of association with efavirenz plasma concentrations in this cohort compared with previous report by Poeta *et al*.38

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**Table 1** Patients’ demographics (median, range) and genotype frequencies.

|  |  |
| --- | --- |
| **Characteristics** | **Pregnancy (n = 77)** |
| Age (years) | 27 (18-39) |
| Weight (kg) | 57 (48-83) |
| Time since diagnosis (months) | 30 (1.5-72) |
| **Pregnancy** |  |
| Gestational age (weeks) | 28 (11-36) |
| First trimester (%) | 5 |
| Second trimester (%) | 25 |
| Third trimester (%) | 70 |
| **Drug regimen and CD4 count** |  |
| TDF/FTC/EFV (%) | 58 |
| 3TC/AZT/EFV (%) | 21 |
| 3TC/TDF/EFV (%) | 21 |
| Duration on regimen (months) | 24 (0.83-45) |
| Baseline CD4 count (cells/mm3) | 294 (5-614) |
| CD4 count change from baseline (cells/mm3) | 235 (21-471) |
| **Genotype frequencies**a |  |
| *CYP2B6* 516G>T (rs3745274) | GG, 0.32; GT, 0.54; TT, 0.14 |
| *CYP2B6* 983T>C (rs28399499) | TT, 0.75; CT, 0.25; CC, 0.00 |

aFull list of genotype frequencies is available in Clin Pharmacol Ther 2015; 97: 298-306 (Ref. 11)

**Table 2** Efavirenzfinal parameter estimates and standard errors obtained from the final population pharmacokinetic model.

|  |  |  |  |
| --- | --- | --- | --- |
| **Parameter** | **Estimate** | **RSE (%)** | **90% CI** |
| CL/FFast (L/h)a | 18  | 9 | (15-21.5) |
| CL/FIntermediate (L/h) | 16.1  | 7 | (15.1-18) |
| CL/FSlow (L/h) | 6.24  | 11 | (4.8-7.5) |
| V/F (L) | 281 | 10 | (241-320) |
| ka (h-1) | 0.61 | 23 | (0.3-0.9) |
|  |  |  |  |
| IIV CL/F (%) | 41.1 | 16 | (36-43) |
| IIV V/F (%) | 26.4 | 33 | (10-40) |
| IIV ka (%) | 86.8 | 21 | (23-107) |
|  |  |  |  |
| *Residual error* |  |  |  |
| Proportional  | 0.085 | 21 | (0.06-0.11) |

aFast, intermediate and slow metabolisers status were based on composite CYP2B6 516 and 983T>C genotype. RSE (%): relative standard error; CL/F: apparent oral clearance; V/F: apparent volume of distribution; ka: absorption rate constant; IIV: inter-individual variability; RSE defined as: (SEestimate/estimate)\*100; CI: confidence interval.

**Table 3** Predicted efavirenz pharmacokinetic parameters in pregnant women stratified by *CYP2B6* metabolic status

|  |  |  |  |
| --- | --- | --- | --- |
| **Dose** | **200 mg** | **400 mg** | **600 mg** |
| **Parametera** | **Slow** | **Intermediate** | **Fast** | **Slow** | **Intermediate** | **Fast** | **Slow** | **Intermediate** | **Fast** |
| AUC0-24 (µg.h/mL) | 26.8 (10.7, 50.4) | 10.4 (3.99, 19.7) | 9.27 (3.52, 21.9) | 53.5 (21.1, 100.9) | 20.8 (8.08, 39.4) | 18.5 (7.11, 35.3) | 79.8 (31.8, 151.7) | 31.1 (12.0, 59.3) | 27.8 (10.7, 53.3) |
| Cmax (µg/mL) | 1.31 (0.56, 2.46) | 0.61 (0.25, 1.16) | 0.57 (0.23, 1.23) | 2.67 (1.03, 4.95) | 1.22 (0.50, 2.31) | 1.13 (0.45, 2.16) | 3.88 (1.55, 7.42) | 1.83 (0.73, 3.49) | 1.71 (0.68, 3.23) |
| Ctrough (µg/mL) | 0.89 (0.33, 1.82) | 0.24 (0.078, 0.51) | 0.20 (0.060, 0.52) | 1.78 (0.66, 3.64) | 0.47 (0.16, 1.02) | 0.39 (0.12, 0.86) | 2.68 (1.00, 5.51) | 0.71 (0.22, 1.53) | 0.59 (0.18, 1.29) |
| C12 (µg/mL) | 1.14 (0.45, 2.10) | 0.44 (0.17, 0.82) | 0.39 (0.15, 0.95) | 2.24 (0.89, 4.18) | 0.87 (0.34, 1.64) | 0.78 (0.30, 1.47) | 3.37 (1.35, 6.31) | 1.31 (0.51, 2.48) | 1.17 (0.45, 2.21) |
| C12 < 1.0 µg/mL (%)b | 41 | 95 | 96 | 7.0 | 61 | 68 | 2.0 | 29 | 38 |
| C12 < 0.47 µg/mL (%)b | 6.4 | 55 | 64 | 1.2 | 13 | 16 | 0 | 3.0 | 6.0 |
| C12 > 4.0 µg/mL (%) | 1.0 | 0 | 0 | 13 | 0.4 | 0.4 | 38 | 1.6 | 1.0 |

aData are presented as mean (90% Confidence Interval); b1.0 µg/mL is the traditional C12 threshold while 0.47 µg/mL is the threshold determined within the ENCORE 1 trial.

**Figure legends**

**Figure 1.** Individual plasma efavirenz concentration-time profiles in pregnant women who received 600 mg efavirenz as part of combination ART. Median C12 was 1.01 µg/mL (range: 0.78-1.26). Horizontal dotted lines show 1.0 µg/mL (upper) and 0.47 µg/mL (lower) and the intersection with the vertical dotted line shows C12.

**Figure 2.** Visual predictive check for the final pharmacokinetic model fitting. The lower, middle and upper dashed lines are the 5th, 50th and 95th percentiles of the observed data, respectively. The shaded regions represent the 90% CIs of the simulated prediction interval of the 5th, 50th, and 95th percentiles. The three sub-groups are represented with symbols: circle, fast metabolisers; triangle, intermidiate metabolisers; exe, slow metabolisers. Goodness of fit plots showing observed versus predicted plasma efavirenz concentration are presented in supplemenatry data (Figure S1).

**Figure 3.** Pharmacokinetic profiles at 200, 400, and 600 mg daily doses. At the 400 mg reduced dose, the mean (90% CI) C12 was 2.24 µg/mL (0.89, 4.18) in pregnant women classified as slow metabolisers, compared with 0.87 µg/mL (0.34, 1.64) in intermediate and 0.78 µg/mL (0.30, 1.47) in fast metabolisers. Further reducing the daily dose to 200 mg resulted in C12 of 1.14 µg/mL (0.45, 2.10), 0.44 µg/mL (0.17, 0.82) and 0.39 µg/mL (0.15, 0.95), respectively. Horizontal dotted lines show 1.0 µg/mL (upper) and 0.47 µg/mL (lower) and the intersection with the vertical dotted line shows C12.

**Figures**

**Figure 1**



**Figure 2**

|  |
| --- |
|  |

**Figure 3**

