**Pharmacogenetics of artemether-lumefantrine influence on nevirapine disposition: clinically significant drug-drug interaction?**

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**Running Title**

PGt of artemether-lumefantrine vs. nevirapine DDI

**Keywords**

Nevirapine, Artemether-Lumefantrine, Drug-Drug Interaction, Pharmacogenetics, CYP2B6

**What is already known about this subject:**

* Single nucleotide polymorphisms in disposition genes partly contributes to variability in nevirapine pharmacokinetics, with *CYP2B6* c.516G>T and c.983T>C reported to increase its exposure.
* Whereas impact of nevirapine on artemether-lumefantrine is well studied, limited studies have investigated influence of artemether-lumefantrine on nevirapine pharmacokinetics and there are no data on the influence of *CYP2B6* c.516G>T and c.983T>C on this interaction.

**What this study adds:**

* This study introduces a novel approach that involved stratifying patients based on their metabolic status for clinical drug-drug interaction studies.
* Irrespective of patient’s *CYP2B6* metabolic status, co-administration with artemether-lumefantrine is unlikely to result in any clinically significant changes in key nevirapine pharmacokinetics. However, the proportion of patients with trough plasma nevirapine concentration below the putative minimum effective concentration threshold doubled in the presence of artemether-lumefantrine, mainly in extensive metabolisers.

**Abstract**

Aims

In this study the influence of first-line antimalarial drug artemether-lumefantrine on the pharmacokinetics of the antiretroviral drug nevirapine was investigated in the context of selected single nucleotide polymorphisms (SNPs) in a cohort of adult HIV-infected Nigerian patients.

Methods

This was a two-period, single sequence crossover study conducted in two stages. In stage 1, 150 HIV-infected patients receiving nevirapine-based antiretroviral regimens were enrolled and genotyped for 7 SNPs. Sparse pharmacokinetic sampling was conducted to identify SNPs independently associated with nevirapine plasma concentration. Patients were categorised as poor, intermediate and extensive metabolisers based on the numbers of alleles of significantly associated SNPs. Intensive sampling was conducted in selected patients from each group. In stage 2, patients received standard artemether-lumefantrine treatment with nevirapine and intensive pharmacokinetic sampling was conducted on day 3.

Results

No clinically significant changes were observed in key nevirapine pharmacokinetic parameters, the 90% confidence interval for the measured changes falling completely within the 0.80-1.25 no-effect boundaries. However, the number of patients with trough plasma nevirapine concentration below the 3,400 ng ml-1 minimum effective concentration increased from 10% without artemether-lumefantrine (all extensive metabolisers) to 21% with artemether-lumefantrine (14% extensive, 4% intermediate, and 3% poor metabolisers).

Conclusions

This approach highlights additional increase in the already existing risk of suboptimal trough plasma concentration, especially in extensive metabolisers when nevirapine is co-administered with artemether-lumefantrine.

**Introduction**

HIV-infection and malaria are major global public health problems together, constituting 10% of global disease burden and responsible for ≥ 2.5 million deaths per year [1,2]. Sub-Saharan Africa has the highest burdens of both diseases [3]. Nigeria is the leading country for malaria deaths worldwide accounting for 29% out of the 78% of the global total of estimated malaria deaths from 15 countries [1]. It also accounts for nearly 50% of all new HIV-infections in sub-Saharan Africa every year and has the second largest HIV epidemic in the world [2,4]. Artemether-lumefantrine is an artemisinin-based combination therapy (ACT) recommended by the World Health Organization (WHO) as a first-line option for the treatment of malaria [5]. Nevirapine on the other hand is a non-nucleoside reverse transcriptase inhibitor (NNRTI) recommended as one of the alternative components of first-line antiretroviral regimens where the preferred first-line options (efavirenz-containing regimens) are contraindicated or not available [6]. Therefore, there are possibilities of co-administering both drugs when HIV-infected patients on nevirapine-based regimens require treatment for malaria.

Hepatic metabolisms of nevirapine and the artemisinins are mainly through cytochrome P450 (CYP) enzymes 3A4 and 2B6 [7-12]. Nevirapine is metabolised to 2- and 12-hydroxynevirapine primarily by CYP3A4 while CYP2B6 is responsible for the formation of 3- and 8-hydroxynevirapine [7,8]. Artemether is demethylated to dihydroartemisinin by CYP3A4 and 2B6 [9-12]. Both nevirapine and the artemether component of artemether-lumefantrine induce CYP3A4 and 2B6 [13,14]. In addition, artemisinins are known to induce CYP2C9, and to both induce and inhibit CYP2B6 *in vitro* thereby making it difficult to accurately predict the net *in vivo* effect [15-17]. Lumefantrine is metabolised by CYP3A4 and inhibits CYP2D6 *in vitro* [18,19]. These create the potential for drug-drug interactions during co-administration of nevirapine with artemether-lumefantrine.

Nevirapine disposition is characterised with wide interindividual variability and more than 80% of this has been attributed to genetic factors, and concomitant medications introduce further variability [20-22]. Sequence variations in nevirapine-disposition genes can alter the pharmacokinetics of nevirapine [23-25]. Single nucleotide polymorphisms (SNPs) in *CYP2B6* have been associated with nevirapine pharmacokinetics and efficacy [26-29]. Additionally, polymorphisms in *CYP3A4*, cytochrome P450 oxidoreductase (*POR*) and peroxisome proliferator-activated receptor alpha (*PPARA*) genes may influence nevirapine disposition [30-33].

Whereas the impact of nevirapine on artemether-lumefantrine pharmacokinetics are well-studied [34-36], data on the influence of artemether-lumefantrine on nevirapine disposition in HIV-infected patients are limited, especially in the context of known pharmacogenetic factors [35,36]. Moreover, while any impact of artemether-lumefantrine on nevirapine exposure may seem unlikely to matter in the context of a single course of artemether-lumefantrine, recrudescence often necessitates taking repeated courses of the antimalarial within short time intervals.

Therefore, the association of selected SNPs in nevirapine disposition genes with its pharmacokinetics, and their influence on changes in its pharmacokinetics when co-administered with artemether-lumefantrine was investigated.

**Methods**

***Patients and Study Design***

Unrelated HIV-infected adult patients were recruited from the Virology Research Clinic, Nigeria (IHVN) Research Clinic, Obafemi Awolowo University Teaching Hospitals Complex (OAUTHC), Ile-Ife. Patients were eligible for the study if they were at least 18 years old, receiving regimens containing 200 mg nevirapine twice daily as part of antiretroviral therapy (ART) for at least 2 months and able to understand the study information. Patients were excluded from the study for any of the following reasons: pregnancy, breastfeeding, concomitant drugs with known or uncertain interaction with nevirapine except the study drugs, severe hepatic or renal dysfunction and hypersensitivity to study drugs. Patients with recent history of poor compliance according to pharmacy ART refill record were also excluded. Participants were enrolled into the study after obtaining written informed consent. Demographic information of each patient was obtained and recorded anonymously to maintain confidentiality. Approval of the study protocol was obtained from OAUTHC Health Research Ethics Committee (HREC) and the study was conducted in accordance with the principles of Declaration of the Helsinki [37].

This was a two-period, single sequence crossover study conducted in two stages. In stage 1, consenting eligible patients were enrolled and genotyped for selected SNPs. Sparse pharmacokinetic sampling was conducted to identify SNPs independently associated with plasma nevirapine concentration. Patients were categorised as poor, intermediate and extensive metabolisers based on the number of alleles of significantly associated SNPs and intensive sampling was conducted in selected patients from each group. In stage 2, patients received standard artemether-lumefantrine treatment with nevirapine and intensive pharmacokinetic sampling was conducted on day 3.

***Drug Administration and Pharmacokinetic Sampling***

In both stages of the study, dried blood spots (DBS) were collected on Whatman® 903 protein saver cards (GE Healthcare, Little Chalfont,Buckinghamshire, UK) from each patient after finger prick with safety lancet. Sparse pharmacokinetic sampling for stage 1 was collected at steady state and at the end of a dosing interval, at 0.5, 1, 2, 3, 4, 6, 8 and 12 h after dose for the intensive pharmacokinetic sampling to obtain baseline nevirapine pharmacokinetic parameters. Patients selected for stage 2 were given a 3-day course of artemether-lumefantrine treatment comprising of 80/480 mg tablets twice daily. Drugs were self-administered at home after meal along with nevirapine-containing ART; only the last dose of artemether-lumefantrine was directly observed at the clinic on the intensive pharmacokinetic sampling day. Samples were collected on up to 5 pre-marked areas on - Whatman® 903 protein saver cards, allowed to air dry for at least 2 h, packed in ziplock bags with desiccants, and stored at room temperature. Samples were transferred to the Department of Molecular and Clinical Pharmacology, University of Liverpool, Liverpool, United Kingdom for SNP genotyping and drug quantification.

***DNA Extraction and SNP Genotyping***

Genomic DNA was extracted from the DBS samples using E.Z.N.A.® Blood DNA Mini Kit (Omega Bio-Tek, Inc., Norcross, GA, USA), according to manufacturer’s protocol. Extracted DNA was quantified using NanoDrop® (Thermo Fisher Scientific Inc., Wilmington, DE, USA) and stored at -20oC until analysis. TaqMan® Genotyping Master Mix and assays for *CYP2B6* c.516G>T (rs3745274; ID: C\_7817765\_60), *CYP2B6* c.785A>G (rs2279343; ID: C\_26823974\_30), *CYP2B6* c.983T>C (rs28399499; ID: C\_60732328\_20), *CYP3A4* c.522-191C>T (rs35599367; ID: C\_59013445\_10), *CYP3A5* c.6986A>G (rs776746; ID: C\_26201809\_30), *POR* c.1508C>T (rs1057868; ID: C\_8890131\_30) and *PPARA* c.208+3819A>G (rs4823613; ID: C\_2985275\_10) SNPs were obtained from Life Technologies Ltd (Paisley, Renfrewshire, UK). Genotyping for the presence of the seven SNPs was performed using previously described and validated real -time PCR assay on a DNA Engine Chromo4 system (Bio-Rad Laboratories, Inc., Hercules, CA, USA) [38]. The PCR protocol involved an initial denaturation step at 95°C for 15 min, followed by 50 cycles of amplification at 95°C for 15 s and final annealing at 60°C for 1 min. Opticon Monitor® version 3.1 (Bio-Rad Laboratories, Inc., Hercules, CA, USA) was used to obtain allelic discrimination plots and make allele calls.

***Quantification of Nevirapine in DBS and Pharmacokinetic Analysis***

Blood volume was assumed from uniform distribution. In addition, a section of each DBS sample was punched into a 7 ml screw cap tube using a 6 mm hole punch before being extracted with 1 ml of 0.1% formic acid in acetonitrile:water (80:20, v/v). A previously described validated liquid chromatography-tandem mass spectrometry (LC-MS/MS) method was used to quantify nevirapine in DBS [39]. Briefly, the assay was validated over the concentration range 50–10000 ng ml-1 with accuracy within 93.3 – 103.5 % and precision within 3.2 – 12.0 %. Plasma concentrations were calculated from DBS concentrations using the formula: Plasma (nevirapine) **=** [{DBS (nevirapine) **/** (1 - HT)} × fbpp] where HT represents patient specific haematocrit value and fbpp is the fraction of nevirapine bound to plasma proteins [40].

Pharmacokinetic analysis of intensive pharmacokinetic data was performed with Kinetica® (version 4.1 InnaPhase Corporation, Philadelphia, PA, USA) using standard non-compartmental approach. Pharmacokinetic parameters, including maximum plasma concentration (Cmax),trough plasma concentration (Ctrough), area under the concentration-time curve (AUC0-12), apparent clearance (CL) and the half-life (t1/2) were obtained from the DBS-derived plasma data.

***Statistical Analysis***

Genotype distributions were tested for compliance with Hardy-Weinberg Equilibrium (HWE) by calculating expected frequencies of genotypes and comparing them to the observed values using Chi Square (χ2)test. Analysis of variance, with post hoc testing using the Tukey test, was used to assess differences in plasma nevirapine concentrations among the genotypes. Statistical significance was defined as P < 0.05.

Linear regression analysis was used to evaluate associations of genetic and non-genetic factors with plasma nevirapine concentrations. Variables with P ≤ 0.10 in univariate analysis were included in multivariate analysis and patients were stratified into three groups for intensive pharmacokinetic sampling using SNPs found to be independently associated. The geometric mean ratio (GMR) with the associated 90% confidence interval (CI) of nevirapine pharmacokinetic parameters in the presence of artemether-lumefantrine (nevirapine + artemether-lumefantrine) to nevirapine alone was computed. Clinically significant interaction was ruled out if the entire 90% CI falls within the no effect boundary of 0.80 to 1.25 [41]. Additionally, the proportion of patients with plasma trough concentrations below 3,400 ng ml-1, a threshold previously associated with virological failure [42,43] is presented to further aid clinical interpretation of the drug-drug interaction data. These analyses were performed using IBM® SPSS Statistics version 20.0 (IBM, Armonk, NY, USA). All figures were produced in GraphPad Prism® version 6.01 (GraphPad Software Inc., San Diego, CA).

**Results**

***Patients’ Demographics***

One hundred and fifty (150) HIV-infected patients on nevirapine-containing regimens were enrolled in the first stage of the study. Patients’ baseline characteristics are summarised in Table 1. The mean (standard deviation) age of the patients was 39.79 (9.66) years, weight was 62.38 (14.32) kg, body mass index (BMI) was 23.57 (4.86) kg/m2 and haematocrit was 0.36 (0.004). Most of the patients were female (72%), Yoruba by tribe (83%) and taking zidovudine and lamivudine (85%) in combination with nevirapine. The average duration on therapy was 3.04 (2.41) years.

***Allele and Genotype Frequencies***

The allele and genotype frequencies of the seven SNPs in the study population are shown in Supplementary Table 1. Comparison of observed vs. expected genotype frequencies showed that all SNPs were in Hardy-Weinberg Equilibrium (*P* > 0.05). There were no homozygote carriers of the rare *CYP2B6* c.983C and *CYP3A4* c.522-191T alleles. *CYP2B6* c.516G>T was strongly correlated with *CYP2B6* c.785A>G (*P* = 1 x 10-7); no apparent associations between other SNPs were observed.

***Association of SNPs in Nevirapine Disposition Genes with its Plasma Concentrations***

Seven patients with DBS nevirapine concentration below the 50 ng ml-1 assay quantification limit were deemed non-adherent and excluded from subsequent analysis. There was considerable inter-individual variability in the DBS-derived plasma nevirapine concentrations ranging from 1,100 ng ml-1 to 13,455 ng ml-1 with geometric mean of 4,884 ng ml-1. The linear regression analysis of the sparse pharmacokinetic data using unstandardized beta regression coefficients (β) as a measure of magnitude of effect size, showed that *CYP2B6* c.983T>C, c.516G>T, and gender were independently associated after correction for multiple testing with β (90% CI) of 1,459 ng ml-1 (675, 2243); 805 ng ml-1 (414, 1197), and -1,298 ng ml-1 (-1923, -673), respectively (Table 2). No significant association was observed with the other SNPs.

Tukey HSD Post Hoc multiple comparisons, indicated significant differences in plasma nevirapine concentrations between *CYP2B6* c.516G>T and c.983T>C genotypes (Figure 1A and C). Also, males had significantly higher nevirapine concentrations than females (P = 0.002) at 5,820 (2,359) vs. 4,563 (2,051) ng ml-1, respectively (Figure 1D).

Therefore, composite *CYP2B6* c.516G>T and c.983T>C were used to stratify patients into three groups based on the total number of variant alleles carried: - extensive metabolisers (EMs, no variant allele), intermediate metabolisers (IMs, a one variant allele) and poor metabolisers (PMs, two or more variant alleles).

***Pharmacokinetics of Nevirapine Alone and with Artemether-Lumefantrine in Extensive Versus Poor Metabolisers***

A total of 30 patients (12 extensive metabolisers, 3 intermediate and 15 poor metabolisers) were included in the intensive pharmacokinetic sampling and they contributed 480 nevirapine concentrations. A poor metaboliser patient was excluded due to non-adherence to the sample collection time points. The plasma concentration-time profiles of nevirapine alone vs. when co-administered with artemether-lumefantrine in all 29 patients, and when stratified based on their metabolic status are presented in Figure 2. The associated pharmacokinetic parameters are presented in Table 3. When the influence of artemether-lumefantrine on nevirapine pharmacokinetics was considered in all the patients, 9% and 7% statistically significant decreases in Cmax and AUC0-12 (P < 0.05) were observed although the changes did not reach the previously established, clinically significant threshold as the 90% CI of GMRs fell within the no effect boundary of 0.80 – 1.25 for both the Cmax [0.91 (0.87 - 0.95)] and AUC0-12 [0.93 (0.90 - 0.96)] when values obtained in the presence of artemether-lumefantrine were compared with the values obtained at baseline (Table 3). Similar comparison, when stratified based on composite *CYP2B6* c.516/983 genotypes, showed that artemether-lumefantrine only resulted in 12% statistically, but not clinically, significant decreases in nevirapine Cmax [0.88 (0.84 - 0.92)] and AUC0-12 [0.88 (0.84 - 0.91)] of EMs. In contrast, nevirapine Cmax [0.94 (0.88, 1.01)] and AUC0-12 [0.97 (0.93, 1.02)] of PMs were unaffected by artemether-lumefantrine.

In the absence of artemether-lumefantrine, Ctrough was below the putative minimum effective concentration (MEC) of 3400 ng ml-1 in 10% of patients, all extensive metabolisers. When co-administered with artemether-lumefantrine, 21% of patients have Ctrough below the MEC. Of these, 14% were extensive metabolisers while 4% and 3% were intermediate and poor metabolisers respectively (Table 3).

**Discussion**

The present study investigated the influence of host genetic polymorphisms on the dynamics of pharmacokinetic interaction between nevirapine and artemether-lumefantrine. The results showed lack of clinically significant interaction in all patients and when stratified according to composite *CYP2B6* c.516/983 genotype. The 90% CI of the GMR of key pharmacokinetic parameters (with the exception of Tmax and t1/2) fell within the no-effect boundaries of 0.80 - 1.25 [41]. Despite evidence that nevirapine disposition is associated with *CYP2B6* SNPs [26-29], and previous reports of > 80% relative genetic contribution to variability in nevirapine exposure [20,21], pharmacogenetic influence of these SNPs on the interaction of artemether-lumefantrine with nevirapine are yet to be reported. The 9% and 7% decreased Cmax and AUC0-12 respectively obtained in pooled analysis, are in agreement with a similar parallel design study in Nigerian patients which reported no significant difference in nevirapine exposure [36]. It however differs in magnitude from the 42% and 46% decreased Cmax and AUC0-12  respectively reported in a previous Ugandan study [35], possibly due to differences in study design (parallel vs. crossover) or sample collection techniques (DBS vs. plasma) and differences in genotype distribution.

Veldkamp *et al* have shown that the proposed plasma nevirapine MEC is ≥ 3,400 ng ml-1 (as depicted in Figure 2) [42,43]. Approximately 14% of EMs had trough plasma nevirapine concentrations below the target concentration in the presence of artemether-lumefantrine compared to 3% of PMs as indicated in Table 3. The small sample size of IMs (n = 3) may be responsible for the nevirapine exposure and other pharmacokinetic parameters of IMs that were outside the no effect range. Inclusion of pharmacogenetics in a drug-drug interaction study is recommended when genetic factor is expected to significantly influence the pharmacokinetics of any of the drugs [44,45]. Composite *CYP2B6* c.516/983 genotype is known to significantly influence nevirapine pharmacokinetics [26-29] as demonstrated in the present study (Supplementary Table 2). Thus, it was anticipated to contribute to the extent of artemether-lumefantrine interaction with nevirapine. Although study results indicated that the decreased nevirapine exposures due to influence of artemether-lumefantrine were mitigated in IMs and PMs, metabolic status was not significantly correlated with the proportion of patients with trough plasma nevirapine concentration below the MEC. The clinical implications of this is that while nevirapine may be considered safe and not requiring dosage adjustment when co-administered with artemether-lumefantrine in the treatment of malaria-HIV co-infected patients irrespective of their metabolic status, caution is warranted most especially in malaria endemic regions. Studies in highly endemic areas have shown repeated episodes of symptomatic malaria within short interval of a prior attack particularly when short acting ACTs like artemether-lumefantrine are used in treatment compared to long acting ACTs [46,47]. This has the potential to decrease efficacy and increase resistance development to nevirapine, considering its low genetic barrier to resistance [48].

Although in vitro inhibition and induction of CYP2B6 by artemether have been reported, in vivo inhibition is unknown and induction is usually the net effect reported [15-17]. Induction of CYP enzymes by artemether seems to be due to activation of the nuclear receptors: constitutive androstane receptor (CAR) and/or pregnane X receptor (PXR) [14,49]. Artemisinin derivatives and their metabolites are reported to differentially affect the activities of CAR isoforms which may be responsible for differences in the inductive capacity between the artemisinin drugs [15,50]. Whereas artemether demonstrated agonist binding to the major isoforms CAR1 and CAR3, dihydroartemisinin acted as weak inverse agonist of CAR1 [50]. Due to the weaker induction capacity of artemether compared to nevirapine, in the presence of nevirapine, artemether is rapidly metabolised by the more potent nevirapine to dihydroartemisinin. Thus, the induction capacity of artemether is largely dependent on dihydroartemisinin, a weaker activator of CAR/PXR dependent gene expression in humans compared to artemether [14,16,50]. Hence, the inability of artemether to consistently induce CYP3A4 and 2B6, coupled with the inverse agonist effect of the dihydroartemisinin metabolite are possible explanations for the small magnitude of decreased nevirapine exposure observed in the present study.

In addition, the extent of CYP induction has been reported to depend upon the baseline levels of CYP enzymes [51]. Because artemether is a substrate and metabolised by CYP2B6, induction of CYP2B6 and 3A4 may be altered in PMs given the likelihood of higher exposure to artemether as a result of decreased clearance compared to EMs. However, low constitutive expression of CYP2B6 in PMs owing to the carriage of variant *CYP2B6* c.516T and/ c.983C alleles implies less enzymes available for induction by the increased amount of artemether, resulting in a small decrease in nevirapine exposure which was almost offset by the increase in exposure due to genetic influence. Hence, the lack of change in nevirapine exposure due to effect of artemether-lumefantrine observed in PMs. On the other hand, the EMs with more CYP2B6 owing to higher expression compared to PMs had greater artemether induction capacity inspite of the probable decreased exposure to artemether to the extent that an appreciable decrease in nevirapine exposure was observed.

Similar non-significant clinical changes in dolutegravir exposures were also reported in a recent study involving ACTs vs. dolutegravir (a second generation integrase strand transfer inhibitor) on healthy Ugandan volunteers [52]. No changes in dolutegravir Cmax and AUC0-24 were reported when co-administered with artemether-lumefantrine, although 42% and 24% significant changes in dolutegravir trough plasma concentration and AUC0-24 respectively were observed with artesunate-amodiaquine [52]. However, genetic influence was not factored into the study. Dolutegravir is primarily metabolised via UGT1A1 [53,54], and functional polymorphisms in the *UGT1A1*, particularly \*28 and \*37 alleles are known to decrease enzyme activity [55]. Thus, possibility of *UGT1A1* polymorphisms influencing dolutegravir exposure due to its intearaction with ACTs can not be ruled out. Hence, the need to incorporate pharmacogenetics in drug interaction studies involving drug(s) with high relative genetic contribution to the pharmacokinetic variation of the drug(s).

Pharmacogenetics is crucial to a better understanding and evaluation of pharmacokinetics as well as magnitude of drug-drug interactions for both new and existing drugs most especially when polymorphic enzymes/proteins are responsible for metabolism or transport of the drugs [44,45]. A major limitation to the clinical implementation of pharmacogenetics is lack of adequate scientific evidence of the clinical relevance of genetics in drug-drug interaction studies of both new and existing drugs. While pharmaceutical industries are expected to provide data for new drugs, the onus is on the academic and research institute to fill the gap for already existing drugs [56]. Furthermore, most pharmacogenetic studies have pharmacokinetic end points with no efficacy or toxicity data integrated. Therefore, better designed-studies incorporating pharmacodynamic data are necessary for the clinical implementation of pharmacogenetics [56]. A major limitation of the present study was the lack of pharmacodynamic end points. In addition, there was lack of certainty on the time since last dose for sparse pharmacokinetic sampling as the last dose was self-administered by patients at home and time of last dose was based on patients’ reports. Furthermore, the study recruited more female than male patients. However, these limitations did not hinder the detection of the influence of composite *CYP2B6* c.516G/983 genotypes as biomarkers to predict the CYP2B6-mediated metabolism of nevirapine, alone and in the presence of artemether-lumefantrine.

In summary, study results demonstrate lack of clinical significance, in the influence of artemether-lumefantrine on nevirapine exposures of both *CYP2B6* c.516/983 EMs and PMs although EMs had greater proportion of patients with trough plasma nevirapine concentration below the putative minimum effective concentration compared to PMs. It also supports efficacy and safety of nevirapine when co-administered with artemether-lumefantrine, irrespective of patients’ metabolic status but caution is warranted in malaria endemic areas.

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**Conflicts of interest**

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**References**

1. WHO. World Health Organization’s World Malaria Report 2015. Geneva, Switzerland. [Online]. Available: http://www.who.int/malaria/publications/world-malaria-report-2015/report/en/ [Accessed 12 October 2016].
2. UNAIDS. UN Joint Programme on HIV/AIDS, Global AIDS Update – 2016, June 2016 [Online]. Available: http://www.refworld.org/docid/574e8d394.html [Accessed 3 April 2017].
3. Chandramohan D, Greenwood BM. Is there an interaction between human immunodeficiency virus and Plasmodium falciparum? Int J Epidemiol 1998; 27:296-301.
4. UNAIDS. UN Joint Programme on HIV/AIDS, Prevention Gap Report, January 2016 [Online]. Available: http://www.refworld.org/docid/57862e014.html [Accessed 15 January 2018].
5. WHO. World Health Organization’s Guideline for the treatment of malaria. Third edition. 2015; Geneva, Switzerland.
6. WHO. World Health Organization Consolidated guidelines on the use of antiretroviral drugs for treating and preventing HIV infection: recommendations for a public health approach. Second edition. 2016; Geneva, Switzerland. [Online]. Available: https://www.ncbi.nlm.nih.gov/books/NBK374294/ [Accessed 24 November 2017].
7. Riska P, Lamson M, MacGregor T, Sabo J, Hattox S, Pav J, *et al.* Disposition and biotransformation of the antiretroviral drug nevirapine in humans. Drug Metabol Dispos1999; 27:895-901.
8. Erickson DA, Mather G, Trager WF, Levy RH, Keirns JJ. Characterization of the in vitro biotransformation of the HIV-1 reverse transcriptase inhibitor nevirapine by human hepatic cytochromes P-450. Drug Metabol Dispos 1999; 27:1488-95.
9. Svensson US, Ashton M. Identification of the human cytochrome P450 enzymes involved in the in vitro metabolism of artemisinin. Br J Clin Pharmacol 1999; 48:528-35.
10. Simonsson US, Jansson B, Hai TN, Huong DX, Tybring G, Ashton M. Artemisinin autoinduction is caused by involvement of cytochrome P450 2B6 but not 2C9. Clin Pharmacol Ther2003; 74:32-43.
11. Navaratnam V, Mansor SM, Sit NW, Grace J, Li Q, Olliaro P. Pharmacokinetics of artemisinin-type compounds. Clin Pharmacokinet 2000; 39:255-70.
12. Honda M, Muroi Y, Tamaki Y, Saigusa D, Suzuki N, Tomioka Y, *et al.* Functional characterization of CYP2B6 allelic variants in demethylation of antimalarial artemether. Drug Metab Dispos 2011; 39:1860-5.
13. Lamson M, MacGregor T, Riska P, Erickson D, Maxfield P, Rowland L, *et al.* Nevirapine induces both CYP3A4 and CYP2B6 metabolic pathways. Clin Pharmacol Ther 1999; 65:137.
14. Burk O, Arnold KA, Nussler AK, Schaeffeler E, Efimova E, Avery BA, *et al*. Antimalarial artemisinin drugs induce cytochrome P450 and MDR1 expression by activation of xenosensors pregnane X receptor and constitutive androstane receptor. Mol Pharmacol 2005; 67:1954-65.
15. Elsherbiny DA, Asimus SA, Karlsson MO, Ashton M, Simonsson US. A model based assessment of the CYP2B6 and CYP2C19 inductive properties by artemisinin antimalarials: implications for combination regimens. J Pharmacokinet Pharmacodyn 2008; 35:203-17.
16. Xing J, Kirby BJ, Whittington D, Wan Y, Goodlett DR. Evaluation of P450 inhibition and induction by artemisinin antimalarials in human liver microsomes and primary human hepatocytes. Drug Metab Dispos 2012; 40:1757-64.
17. Ericsson T, Masimirembwa C, Abelo A, Ashton M. The evaluation of CYP2B6 inhibition by artemisinin antimalarials in recombinant enzymes and human liver microsomes.Drug Metab Lett 2012; 6:247-57.
18. Lefèvre G, Carpenter P, Souppart C, Schmidli H, McClean M, Stypinski D. Pharmacokinetics and electrocardiographic pharmacodynamics of artemether-lumefantrine (Riamet) with concomitant administration of ketoconazole in healthy subjects. Br J Clin Pharmacol 2002; 54:485-92.
19. Kiang TK, Wilby KJ, Ensom MH. Clinical pharmacokinetic drug interactions associated with artemisinin derivatives and HIV-antivirals. *Clin Pharmacokinet* 2014; 53:141-153.
20. Micheli JE, Chinn LW, Shugarts SB, Patel A, Martin JN, Bangsberg DR *et al*. Measuring the overall genetic component of nevirapine pharmacokinetics and the role of selected polymorphisms: towards addressing the missing heritability in pharmacogenetic phenotypes? Pharmacogenet Genomics 2013; 23: 591–6.
21. Siccardi M, Olagunju A, Simiele M, D’Avolio A, Calcagno A, Di Perri G, *et al*. Class-specific relative genetic contribution for key antiretroviral drugs. J Antimicrob Chemother2015; 70:3074–79.
22. Stöhr W, Back D, Dunn D, Sabin C, Winston A, Gilson R, *et al*. Factors influencing efavirenz and nevirapine plasma concentration: effect of ethnicity, weight and co-medication. Antivir Ther 2008; 13:675-85.
23. Lang T, Klein K, Fischer J, Nüssler AK, Neuhaus P, Hofmann U, *et al*. Extensive genetic polymorphism in the human CYP2B6 gene with impact on expression and function in human liver. Pharmacogenetics 2001; 11: 399–415.
24. Lamba V, Lamba J, Yasuda K, Strom S, Davila J, Hancock ML, *et al*. Hepatic CYP2B6 expression: gender and ethnic differences and relationship to CYP2B6 genotype and CAR (constitutive androstane receptor) expression. J Pharmacol Exp Ther 2003; 307:906–22.
25. Klein K, Lang T, Saussele T, Barbosa-Sicard E, Schunck WH, Eichelbaum M, *et al*. Genetic variability of CYP2B6 in populations of African and Asian origin: allele frequencies, novel functional variants, and possible implications for anti-HIV therapy with efavirenz. Pharmacogenet Genomics 2005; 15:861–73.
26. Rotger M, Colombo S, Furrer H, Bleiber G, Buclin T, Lee BL, *et al*. Influence of CYP2B6 polymorphism on plasma and intracellular concentrations and toxicity of efavirenz and nevirapine in HIV-infected patients. Pharmacogenet Genomics 2005; 15(1):1–5.
27. Penzak SR, Kabuye G, Mugyenyi P, Mbamanya F, Natarajan V, Alfaro RM, *et al*. Cytochrome P450 2B6 (CYP2B6) G516T influences nevirapine plasma concentrations in HIV-infected patients in Uganda. HIV Med 2007; 8: 86–91.
28. Saitoh A, Sarles E, Capparelli E, Aweeka F, Kovacs A, Burchett SK, *et al.* CYP2B6 genetic variants are associated with nevirapine pharmacokinetics and clinical response in HIV-1-infected children. AID*S* 2007; 21: 2191-9.
29. Mahungu TW, Smith CJ, Turner F, Egan D, Youle M, Johnson MA, *et al*. Cytochrome P450 2B6 516G>T is associated with plasma concentrations of nevirapine at both 200 mg twice daily and 400 mg once daily in an ethnically diverse population. HIV Med 2009; 10:310-7.
30. Lamba JK, Lin YS, Schuetz EG, Thummel KE. Genetic contribution to variable human CYP3A-mediated metabolism. Adv Drug Deliv Rev 2002; 54:1271-94.
31. Agrawal V, Choi JH, Giacomini KM, Miller WL. Substrate specific modulation of CYP3A4 activity by genetic variants of cytochrome P450 oxidoreductase. Pharmacogenet Genomics 2010; 20: 611–8.
32. de Jonge H, Metalidis C, Naesens M, Lambrechts D, Kuypers DR. The P450 oxidoreductase \*28 SNP is associated with low initial tacrolimus exposure and increased dose requirements in CYP3A5-expressing renal recipients. Pharmacogenomics 2011; 12:1281–91.
33. Klein K, Thomas M, Winter S, Nussler AK, Niemi M, Schwab M, *et al*. PPARA: a novel genetic determinant of CYP3A4 in vitro and in vivo. Clin Pharmacol Ther 2012; 91:1044-52.
34. Kredo T, Mauff K, van der Walt JS, *et al.* Interaction between artemether-lumefantrine and nevirapine-based antiretroviral therapy in HIV-1-infected patients. Antimicrob Agents Chemother 2011; 55:5616–23.
35. Byakika-Kibwika P, Lamorde M, Mayito J, Nabukeera L, Namakula R, Mayanja-Kizza H, *et al.* Significant pharmacokinetic interactions between artemether/lumefantrine and efavirenz or nevirapine in HIV-infected Ugandan adults. J Antimicrob Chemother 2012; 67:2213–21.
36. Parikh S, Fehintola F, Huang L, Olson A, Adedeji WA, Darin KM, *et al*. Artemether-lumefantrine exposure in HIV-infected Nigerian subjects on nevirapine-containing antiretroviral therapy. Antimicrob Agents Chemother2015; 59:7852-56.
37. WMA. World Medical Association Declaration of Helsinki-Ethical Principles for Medical Research Involving Human Subjects. [Online]. Available: https://www.wma.net/policies-post/wma-declaration-of-helsinki-ethical-principles-for-medical-research-involving-human-subjects/ (2013). [Accessed 5 April 2017]
38. Olagunju A, Khoo S, Owen A. Pharmacogenetics of nevirapine excretion into breast milk and infants' exposure through breast milk versus postexposure prophylaxis. Pharmacogenomics 2016; 17:891-906.
39. Olagunju A, Amara A, Waitt C, Else L, Penchala SD, Bolaji O, *et al*. Validation and clinical application of a method to quantify nevirapine in dried blood spots and dried breast-milk spots. J Antimicrob Chemother 2015; 70:2816-22.
40. Kromdijk W, Mulder JW, Rosing H, Smit PM, Beijnen JH, Huitema ADR. Use of dried blood spots for the determination of plasma concentrations of nevirapine and efavirenz. J Antimicrob Chemother 2012; 67:1211–16.
41. Prueksaritanont T, Chu X, Gibson C, Cui D, Yee KL, Ballard J, *et al*. Drug–Drug Interaction Studies: Regulatory Guidance and An Industry Perspective. AAPS J 2013; 15:629-45.
42. Veldkamp AI, Weverling GJ, Lange JM, Montaner JS, Reiss P, Cooper DA, *et al*. High exposure to nevirapine in plasma is associated with an improved virological response in HIV-1-infected individuals. AIDS 2001; 15:1089-95.
43. Kappelhoff BS, Crommentuyn KM, de Maat MM, Mulder JW, Huitema AD, Beijnen JH. Practical guidelines to interpret plasma concentrations of antiretroviral drugs. Clin Pharmacokinet 2004; 43:845-53.
44. European Medicine Agency. 2012. Guideline on the use of pharmacogenetic methodologies in the pharmacokinetic evaluation of medicinal products (EMA/CHMP/37646/2009. London. [Online]. Available: [www.ema.europa.eu/docs/en\_GB/document\_library/Scientific\_guideline/2012/02/WC500121954.pdf](http://www.ema.europa.eu/docs/en_GB/document_library/Scientific_guideline/2012/02/WC500121954.pdf) (Accessed 18 May 2018).
45. Food and Drug Administration. 2013. Clinical pharmacogenomics: premarket evaluation in early-phase clinical studies and recommendations for labeling. [Online]. Silver Spring. Available at https://www.fda.gov/downloads/Drugs/GuidanceComplianceRegulatoryInformation/Guidances/UCM337169.pdf (Accessed 20 May 2018).
46. Sagara I, Fofana B, Gaudart J, Sidibe B, Togo A, Toure S, *et al*. Repeated artemisinin-based combination therapies in a malaria hyperendemic area of Mali: efficacy, safety, and public health impact. Am J Trop Med Hyg 2012; 87:50-6.
47. Cairns ME, Walker PG, Okell LC, Griffin JT, Garske T, Asante KP, et al. Seasonality in malaria transmission: implications for case-management with long-acting artemisinin combination therapy in sub-Saharan Africa. Malar J 2015; 14:321.
48. Llibre JM, Schapiro JM, Clotet B. Clinical implications of genotypic resistance to the newer antiretroviral drugs in HIV-1-infected patients with virological failure. Clin Infect Dis. 2010; 50:872-81.
49. Simonsson US, Lindell M, Raffalli-Mathieu F, Lannerbro A, Honkakoski P, Lang MA. In vivo and mechanistic evidence of nuclear receptor CAR induction by artemisinin. Eur J Clin Invest 2006; 36:647-53.
50. Burk O, Piedade R, Ghebreghiorghis L, Fait JT, Nussler AK, Gil JP, *et al*. Differential effects of clinically used derivatives and metabolites of artemisinin in the activation of constitutive androstane receptor isoforms. Br J Pharmacol 2012; 167:666-81.
51. Lin JH. CYP induction-mediated drug interactions: in vitro assessment and clinical implications. Pharm Res 2006; 23:1089-116.
52. Walimbwa SI, Lamorde M, Waitt C, Kaboggoza J, Else L, Byakika-Kibwika P, *et al*. Dolutegravir interactions with artemether-lumefantrine and amodiaquine-artesunate. [Abstract #459]. Conference on Retroviruses and Opportunistic Infections (CROI 2018). Hynes Convention Centre, Boston, MA, USA, 04-07 March 2018.
53. Castellino S, Moss L, Wagner D, Borland J, Song I, Chen S, *et al*. Metabolism, excretion, and mass balance of the HIV-1 integrase inhibitor dolutegravir in humans. Antimicrob Agents Chemother. 2013; 57:3536-46.
54. Chen S, St Jean P, Borland J, Song I, Yeo AJ, Piscitelli S, *et al*. Evaluation of the effect of UGT1A1 polymorphisms on dolutegravir pharmacokinetics. Pharmacogenomics. 2014; 15:9-16.
55. Hall D, Ybazeta G, Destro-Bisol G, Petzl-Erler ML, Di Rienzo A. Variability at the uridine diphosphate glucuronosyltransferase 1A1 promoter in human populations and primates. Pharmacogenetics. 1999; 9:591-9.
56. Swen JJ, Huizinga TW, Gelderblom H, de Vries EG, Assendelft WJ, Kirchheiner J, *et al*. Translating pharmacogenomics: challenges on the road to the clinic. PLoS Med. 2007; 4(8):e209.

**Figure Legends**

**Figure 1.** Scatter plots showing significant differences in plasma nevirapine concentrations between patients based on*CYP2B6* c.516G>T genotypes (A), *CYP2B6* c.785A>G genotypes (B), *CYP2B6* c.983T>C genotypes (C) and Gender (D).

**Figure 2.** Plasma concentration-time profiles of nevirapine baseline (NVP) and in the presence of artemether-lumefantrine (NVP + AL) in all the patients and when stratified based on composite *CYP2B6* c.516/983 (A), extensive metabolisers (EMs) (B), intermediate metabolisers (IMs) (C) and poor metabolisers (PMs) (D). Abbreviations: AUC0-12 during a 12-hour dosing interval, area under the concentration-time curve; Cmax, maximum plasma concentration; MEC, minimum effective concentration (3,400 ng ml-1); NVP, nevirapine.

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| **Table 1.** Demographics of HIV-infected patients at the time of sampling | |
| **Characteristics** | **Statistics** |
| Age (years) | 39.79 ± 9.66 |
| Weight (kg) | 62.38 ± 14.32 |
| BMI (kg/m2) | 23.57 ± 4.86 |
| Haematocrit value | 0.36 ± 0.04 |
| Gender, n (%) |  |
| Female | 108 (72) |
| Male | 42 (28) |
| Ethnicity, n (%) |  |
| Hausa | 9 (6) |
| Igbo | 16 (11) |
| Yoruba | 125 (83) |
| NRTI backbone, n (%) |  |
| ABC/3TC | 3 (2) |
| AZT/3TC | 128 (85) |
| TDZ/3TC | 19 (13) |
| Cotrimoxazole prophylaxis, n (%) |  |
| Yes | 33 (22) |
| No | 117 (78) |
| Duration on ART (years) | 3.04 ± 2.41 |
| Data presented as mean ± standard deviation, unless otherwise indicated. Abbreviations: n, sample size; NRTI, nucleoside reverse transcriptase inhibitors; ABC, abacavir; AZT, zidovudine; TDF, tenofovir disoproxil fumarate; CTX, cotrimoxazole. | |

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| **Table 2.** Linear regression analysis of factors influencing plasma nevirapine concentration | | | | | |
| **Predictors** | **Univariate** | | **Multivariate** | | |
| β (ng ml-1) (90 % CI) | P-value | β (ng ml-1) (90 % CI) | P-value | |
| Age (years) | 16 (-15, 470) | 0.402 |  |  |  |
| BMI (kg/m2) | 6 (-57, 68) | 0.880 |  |  |  |
| SNPs |  |  |  |  |  |
| *CYP2B6* c.516G>T | 711 (298, 1124) | 0.005 | 805 (414, 1197) | 0.001 | 0.014\* |
| *CYP2B6* c.785A>G | 656 (237, 1076) | 0.011 |  |  |  |
| *CYP2B6* c.983T>C | 1179 (346, 2012) | 0.020 | 1459 (675, 2243) | 0.002 | 0.028\* |
| *CYP3A4* c.522-191C>T | -237 (-2094, 1621) | 0.833 |  |  |  |
| *CYP3A5* c.6986A>G | 57 (-586, 700) | 0.883 |  |  |  |
| *POR* c.1508C>T | -213 (-866, 440) | 0.589 |  |  |  |
| *PPARA* c.208+3819A>G | -151 (-585, 284) | 0.566 |  |  |  |
| Gender | -1234 (-1894, -574) | 0.002 | -1298 (-1923, -673) | 0.001 | 0.014\* |
| Ethnicity | -616 (-1186, -45) | 0.076 |  |  |  |
| NRTI backbone | 104 (-727, 935) | 0.837 |  |  |  |
| CTX prophylaxis | 55 (-645, 754) | 0.897 |  |  |  |
| Duration on ART | 51 (-79, 181) | 0.517 |  |  |  |
| \*Bonferroni corrected P-value for multiple comparison; Abbreviations: ART, antiretroviral therapy; β, unstandardised beta coefficient; CI, confidence interval; CTX, cotrimoxazole; CYP, cytochrome P450 enzyme; NRTI, nucleoside reverse transcriptase inhibitors; PPARA, peroxisome proliferator-activated receptor alpha; POR, cytochrome P450 oxidoreductase; SNP, single nucleotide polymorphism. | | | | | |

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| **Table 3.** Influence of artemether-lumefantrine (AL) on nevirapine (NVP) pharmacokinetics in all patients and when stratified as extensive and poor metabolisers | | | |
| **PK parameters** | **NVP alone** | **NVP + AL** | **GMR (90% CI)\*** |
| All Patients (n = 29) | |
| Ctrough (ng ml-1) | 5 071 (2 468) | 4 832 (2 494) | 0.95 (0.91 - 0.99) |
| Cmax (ng ml-1) | 7 770 (2 992) | 7 064 (2 973) | 0.91 (0.87 - 0.95) |
| Tmax (h) | 1.83 (1.16) | 1.71 (1.06) | 0.94 (0.74 - 1.13) |
| AUC0-12 (ng**·**h ml-1) | 72 312 (29 022) | 67 025 (31 115) | 0.93 (0.90 - 0.96) |
| t1/2 (h) | 18 (8) | 21 (8) | 1.17 (1.02 - 1.31) |
| CL/F (l h-1) | 1.96 (1.23) | 1.89 (1.51) | 0.97 (0.85 - 1.10) |
| Ctrough < MEC (%) | 10% | 21 |  |
|  | Extensive Metabolisers (n = 12) | |  |
| Ctrough (ng ml-1) | 3 813 (1 222) | 3 580 (1 219) | 0.94 (0.88 - 1.02) |
| Cmax (ng ml-1) | 6 323 (1 644) | 5 558 (1 439) | 0.88 (0.84 - 0.92) |
| Tmax (h) | 1.97 (1.14) | 1.66 (1.11) | 0.84 (0.52 - 1.35) |
| AUC0-12 (ng**·**h ml-1) | 57 729 (16 525) | 50 630(14 567) | 0.88 (0.84 - 0.91) |
| t1/2 (h) | 14 (4) | 18 (8) | 1.29 (1.04 - 1.66) |
| CL/F (l h-1) | 3.01 (1.18) | 2.82 (1.65) | 0.94 (0.78 - 1.12) |
| Ctrough < MEC (%) | 10% | 14 |  |
|  | Intermediate Metabolisers (n = 3) | |  |
| Ctrough (ng ml-1) | 5 032 (950) | 4 664 (1 514) | 0.93 (0.71 - 1.20) |
| Cmax (ng ml-1) | 7 307 (1902) | 6 483 (1 052) | 0.89 (0.73 - 1.08) |
| Tmax (h) | 2.08 (1.15) | 2.52 (1.15) | 1.21 (0.47 - 3.10) |
| AUC0-12 (ng**·**h ml-1) | 71 139 (16 029) | 65 539 (13 477) | 0.92 (0.75 - 1.13) |
| t1/2 (h) | 20 (1) | 18 (8) | 0.90 (0.40 - 2.17) |
| CL/F (l h-1) | 1.87 (0.38) | 2.07 (1.32) | 1.11 (0.55 - 2.24) |
| Ctrough < MEC (%) | - | 4 |  |
|  | Poor Metabolisers (n = 14) | |  |
| Ctrough (ng ml-1) | 6 486 (2 722) | 6 297 (2 700) | 0.97 (0.92 - 1.03) |
| Cmax (ng ml-1) | 9 394 (3 297) | 8 838 (3 247) | 0.94 (0.88 - 1.01) |
| Tmax (h) | 1.67 (1.26) | 1.62 (1.03) | 0.97 (0.67 - 1.42) |
| AUC0-12 (ng**·**h ml-1) | 88 018 (31 868) | 85 655 (33 820) | 0.97 (0.93 - 1.02) |
| t1/2 (h) | 22 (9) | 24 (8) | 1.10 (0.95 - 1.28) |
| CL/F (l h-1) | 1.37 (0.83) | 1.32 (1.03) | 0.97 (0.83 - 1.12) |
| Ctrough < MEC (%) | - | 3 |  |
| All data presented as geometric mean (standard deviation). Abbreviations: AL, artemether-lumefantrine; AUC0-12, area under the plasma concentration-time curve during a 12-hour dosing interval; CI, confidence interval; CL/F, apparent clearance; Cmax, maximum plasma concentration; Ctrough, minimum plasma concentration; GMR, geometric mean ratio [i.e. (NVP + AL)/NVP (baseline)]; MEC, minimum effective concentration; n, sample size; NVP, nevirapine; t1/2, half-life; Tmax, time to reach maximum plasma concentration. | | | |



Figure 1: Scatter dot plots indicating significant differences between plasma nevirapine concentrations of A) *CYP2B6* c.516G>T genotypes, B) *CYP2B6* c.785A>G genotypes, C) *CYP2B6* c.983T>C genotypes and D) Gender.



Figure 2: Plasma concentration-time profiles of nevirapine baseline (NVP) and in the presence of artemether-lumefantrine (NVP + AL) in A) all the patients and when stratified based on composite *CYP2B6* c.516/983 B) extensive (EMs), C) intermediate (IMs) and D) poor metabolisers (PMs). MEC: - minimum effective concentration (3,400 ng ml-1).