

Effect of dihydroartemisinin/piperazine for malaria intermittent preventive treatment on dolutegravir exposure in pregnant women living with HIV

Clifford G. Banda ^{1,2,3}, Dumisile Nkosi¹, Elizabeth Allen^{2,4}, Lesley Workman^{2,4}, Mwayiwawo Madanitsa^{5,6}, Marumbo Chirwa¹, Mayamiko Kapulula⁵, Sharon Muyaya⁵, Steven Munharo⁵, Lubbe Wiesner ², Kamija S. Phiri^{3,5}, Victor Mwapasa³, Feiko O. Ter Kuile⁷, Gary Maartens ² and Karen I. Barnes ^{2,4*}

¹Malawi-Liverpool-Wellcome Trust Clinical Research Programme, Blantyre, Malawi; ²Division of Clinical Pharmacology, Department of Medicine, University of Cape Town, Cape Town, South Africa; ³Kamuzu University of Health Sciences, Blantyre, Malawi (formerly College of Medicine and Kamuzu College of Nursing, University of Malawi); ⁴WorldWide Antimalarial Resistance Network (WWARN), Pharmacology Scientific Group, University of Cape Town, Cape Town, South Africa; ⁵Training and Research Unit of Excellence, Blantyre, Malawi; ⁶Department of Clinical Sciences, Malawi University of Science and Technology, Limbe, Malawi; ⁷Department of Clinical Sciences, Liverpool School of Tropical Medicine, Liverpool, UK

*Corresponding author. E-mail: karen.barnes@uct.ac.za

Received 25 August 2021; accepted 18 February 2022

Background: In sub-Saharan Africa, the burdens of malaria and HIV infections overlap. In settings with moderate-to-high malaria transmission intensity, pregnant women living with HIV (PLWH) require both ART and malaria intermittent preventive treatment (IPTp). Dihydroartemisinin/piperazine has been identified as a promising alternative to sulfadoxine/pyrimethamine for IPTp. However, another antimalarial drug, artesunate/amodiaquine, similar to dihydroartemisinin/piperazine, was previously shown to reduce dolutegravir exposure in non-pregnant adults.

Objectives: To investigate the effect of dihydroartemisinin/piperazine on dolutegravir plasma exposure in pregnant women on dolutegravir-based ART.

Methods: We conducted an open-label, non-randomized, fixed-sequence, pharmacokinetic study in PLWH in Malawi. Dolutegravir concentrations were measured over a 24 h period, before and after the recommended 3 day treatment dose of dihydroartemisinin/piperazine in 12 pregnant women in their second or third trimester. Non-compartmental analysis was performed, and geometric mean ratios (GMRs) and 90% CIs were generated to compare dolutegravir pharmacokinetic parameters between the two treatment periods.

Results: Co-administration of dihydroartemisinin/piperazine and dolutegravir increased dolutegravir's overall exposure (AUC_{0-24}) and C_{max} by 30% (GMR 1.30; 90% CI 1.11–1.52) and 31% (GMR 1.31; 90% CI 1.13–1.51), respectively. The dolutegravir trough (C_{24}) concentration increased by 42% (GMR 1.42; 90% CI 1.09–1.85). The combined treatments were well tolerated with no serious adverse events observed.

Conclusions: Dihydroartemisinin/piperazine may be administered with dolutegravir-based ART in pregnant women as the modest increase in dolutegravir exposure, similar to pharmacokinetic parameter values published previously, ensures its efficacy without any clinically significant adverse events observed in this small study.

Introduction

In sub-Saharan Africa, malaria and HIV infections are endemic.¹ Pregnant women who are infected with malaria are at increased risk of adverse outcomes including maternal anaemia, severe malaria and low-birth-weight and stillbirth deliveries, with these

adverse outcomes more frequent in pregnant women living with HIV (PLWH).² Preventing malaria in PLWH is, therefore, a key priority.

The WHO recommends intermittent preventive therapy during the second and third trimesters of pregnancy (IPTp) in moderate-to-high intensity malaria transmission settings. This

involves administering standard antimalarial treatment doses regularly during pregnancy. In pregnant women who are not living with HIV, the WHO recommends the sulphonamide-based therapy sulfadoxine/pyrimethamine (SP). However, the efficacy of SP is being undermined by increasing SP resistance.³ PLWH cannot receive SP for IPTp if they are taking another sulphonamide-based combination, trimethoprim/sulfamethoxazole (co-trimoxazole), to prevent opportunistic infections. Therefore, IPTp with dihydroartemisinin/piperazine is being investigated as add-on therapy to co-trimoxazole in PLWH.^{4,5}

Dihydroartemisinin/piperazine is an artemisinin-based combination therapy (ACT), comprising dihydroartemisinin for rapidly suppressing any existing parasite load and piperazine for clearing any remaining parasites and providing protection against new infections, given its long elimination half-life ($t_{1/2}$).⁶ In non-pregnant Ugandan adults, the related ACT artesunate/amodiaquine was found to reduce dolutegravir exposure by 24% and trough concentrations by 42%,⁷ although these remained above the purported minimum efficacious dolutegravir concentration of 300 ng/mL,⁸ the mechanism of this interaction is unknown. Piperazine, like amodiaquine, is a 4-aminoquinoline,⁹ and may thus also reduce dolutegravir exposure. We assessed whether a 3 day treatment course of dihydroartemisinin/piperazine altered plasma dolutegravir exposure in PLWH in Malawi.

Materials and methods

Study design, pharmacokinetic sampling and assay

An open-label, non-randomized, fixed-sequence, pharmacokinetic study was conducted between December 2019 and July 2020 in PLWH in Malawi. Consenting pregnant women on efavirenz-based ART were switched to a dolutegravir-based ART regimen from 16 weeks of gestation. They remained on this new regimen for 4 weeks (Sequence 1) to allow efavirenz enzyme induction effects to wane. The first dihydroartemisinin/piperazine dose was observed, as were doses of dolutegravir-based ART and dihydroartemisinin/piperazine on the last (third) day of dihydroartemisinin/piperazine treatment. Blood samples for dolutegravir concentrations were collected at the same timepoints before (Sequence 2) and after (Sequence 3) co-administration of dolutegravir-based ART and dihydroartemisinin/piperazine (Figure S1, available as [Supplementary data](#) at JAC Online). Dolutegravir plasma concentrations were quantified using LC-MS/MS at the University of Cape Town, South Africa. For further details, see Texts S1 and S2. Routine antenatal care continued in parallel with all study procedures.

A sample size of 14 participants was calculated to have 80% power to detect a change in AUC outside the FDA limits for bioequivalence.¹⁰ Assuming 10% loss to follow-up, 16 participants were planned for recruitment. The study was registered on PACTR.samrc.ac.za (PACTR201910580840196) and research ethics committee approval was granted by the Malawi College of Medicine, University of Cape Town, and Liverpool School of Tropical Medicine, UK.

Safety assessments

Participants were followed up from enrolment until 28 days after dihydroartemisinin/piperazine co-administration. At scheduled and unscheduled visits, symptom-directed history and examination were conducted. All adverse effects were graded and independently assessed by two physician investigators for any relationship to co-administration of dihydroartemisinin/piperazine and dolutegravir-based ART. HIV viral load was measured prior to, and repeated 28 days after,

dihydroartemisinin/piperazine dosing to assess whether participants remained virologically suppressed.

Statistical analysis

Data from Sequences 2 and 3 were analysed using non-compartmental analysis. Pharmacokinetic parameters were estimated for AUC to 24 h post-dose (AUC_{0-24}), $t_{1/2}$, C_{max} and T_{max} . Dolutegravir's CL/F was calculated using the equation $CL/F = \text{dose}/AUC_{0-24}$, while trough concentrations (C_{24}) were estimated from the sample collected just before the next dolutegravir dose. Pharmacokinetic data were log-transformed to calculate the geometric mean ratio (GMR) and 90% CIs of Sequence 3 to Sequence 2 parameters. Changes in pharmacokinetic parameters between the two sequences were considered statistically significant when the 90% CI of the GMR did not cross the value of 1.¹⁰ Since isoniazid is known to inhibit CYP3A4,¹¹ a minor metabolic pathway for dolutegravir, *post hoc* analyses were stratified by concomitant isoniazid use to explore any impact on plasma dolutegravir exposure. Analyses were performed using Stata version 15.1.

Results

Study profile

Twenty pregnant women were screened for eligibility and 13 were recruited. Twelve participants had quantifiable plasma dolutegravir concentrations at all required timepoints (Figure S2). The median (range) gestational age during Sequence 2 was 28 (24–33) weeks. Four participants were receiving TB prophylaxis with isoniazid and pyridoxine, initiated when they started ART, in line with the national HIV treatment policy at that time (Table S1).

Pharmacokinetics of dolutegravir

Dolutegravir exposure was increased when co-administered with dihydroartemisinin/piperazine, with the AUC_{0-24} and C_{max} increased by 30% (GMR 1.30; 90% CI 1.11–1.52) and 31% (GMR 1.31; 90% CI 1.13–1.51), respectively. Dolutegravir C_{24} values were 42% higher (GMR 1.42; 90% CI 1.09–1.85) during co-administration with dihydroartemisinin/piperazine (Figure 1 and Table 1).

Impact of isoniazid prophylaxis

The four participants on isoniazid had higher dolutegravir exposure at baseline (Table S2). In the eight participants not on isoniazid, dolutegravir exposure increased by 38% (GMR 1.38; 90% CI 1.13–1.70) following co-administration with dihydroartemisinin/piperazine. However, in those on isoniazid, dolutegravir exposure remained similar when dihydroartemisinin/piperazine was co-administered with dolutegravir-based ART (GMR 1.15; 90% CI 0.93–1.43).

Treatment-emergent adverse events and viral load changes

A total of 16 adverse events occurred in the 13 participants enrolled. All these events were assessed as mild, with the exception of one incident of catheter site pain, which was assessed to be of moderate severity as paracetamol was given. Two events were suspected to be associated with co-administration of dihydroartemisinin/piperazine and dolutegravir-based ART: nausea that

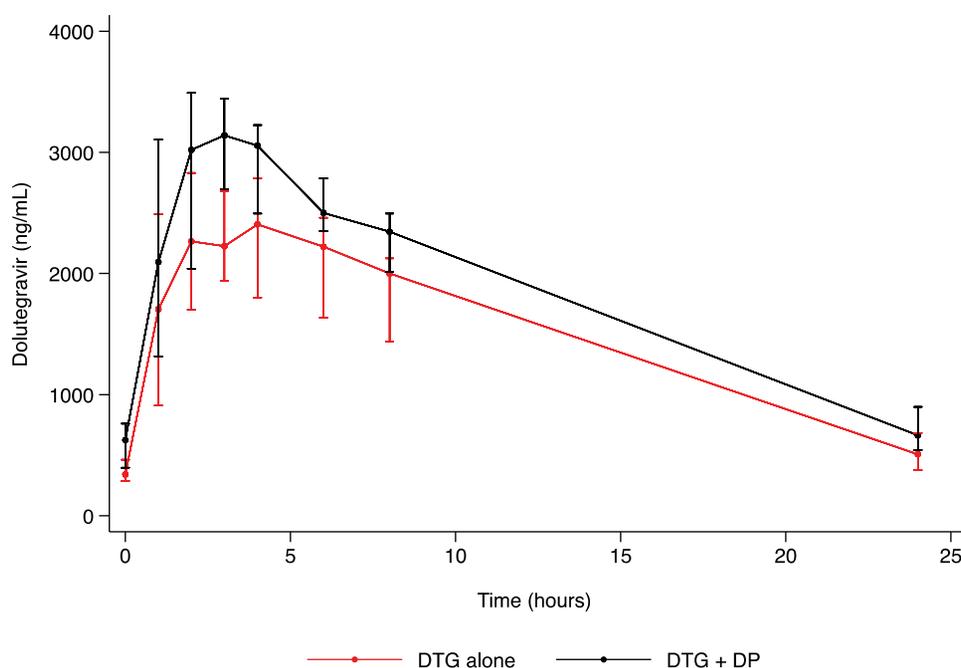


Figure 1. Plasma dolutegravir (DTG) concentration–time profile following administration of DTG-based ART alone (red line) and with a treatment course of dihydroartemisinin/piperazine (+ DP; black line) in 12 pregnant women. Data are presented as median (IQR). This figure appears in colour in the online version of JAC and in black and white in the print version of JAC.

developed 10 min after drug administration and resolved within 1 h of onset, and a pruritic rash that started 2 h after co-administration of study drugs and resolved within 1 day of onset (Table S3). Neither of these events were associated with higher dolutegravir exposure. In all 13 participants enrolled, viral load remained below 50 copies/mL throughout the study.

Discussion

We investigated the impact of the promising antimalarial for IPTp, dihydroartemisinin/piperazine, on the pharmacokinetic profile of dolutegravir when co-administered in PLWH. Dihydroartemisinin/piperazine modestly increased dolutegravir's overall exposure, resulting in pharmacokinetic parameter values similar to those published previously, without any clinically significant adverse events observed in this small study. These findings are reassuring and suggest that dihydroartemisinin/piperazine IPTp can be safely administered with dolutegravir-based ART without reducing dolutegravir exposure (as previously reported with the related antimalarial amodiaquine⁷).

The mechanism behind the increased dolutegravir exposure when co-administered with dihydroartemisinin/piperazine is unclear, but this could be driven by improved bioavailability or reduced clearance. Both piperazine and dolutegravir are substrates of cytochrome P450 3A4 (CYP3A4) enzymes.¹² However, inhibition of CYP3A4 is an unlikely mechanism as piperazine is not known to inhibit CYP3A4, and strong inhibitors of CYP3A4, e.g. cobicistat, do not increase dolutegravir exposure significantly.¹³ We hypothesize that the increased dolutegravir exposure

could be due to dihydroartemisinin/piperazine inhibiting efflux transporters involved in dolutegravir clearance. Dolutegravir is a substrate of P-glycoprotein and breast cancer resistance protein (BCRP) transporters.¹⁴ Mefloquine, an aminoquinoline similar to piperazine, inhibits P-glycoprotein;¹⁵ thus, this is a plausible mechanism. *In vitro* mechanistic studies are warranted.

Dolutegravir exposure, when administered with dihydroartemisinin/piperazine, falls within a range previously described in pharmacokinetic studies during pregnancy.^{16,17} In these studies, dolutegravir exposure was well tolerated, suggesting that the modest increase observed in our study is unlikely to be associated with safety concerns. Furthermore, this increase could help ensure dolutegravir's efficacy; C_{24} values during co-administration with dihydroartemisinin/piperazine were above the purported minimum effective concentration of 300 ng/mL in all study participants.⁸

Our study has limitations. Firstly, due to the small sample size, we were unable to accurately assess the impact of isoniazid on dolutegravir exposure when co-administered with dihydroartemisinin/piperazine; adequately powered, prospective pharmacokinetic studies are needed to investigate this question. Secondly, our participants received dihydroartemisinin/piperazine for IPTp, as part of another study (PACTR201910 580840196),¹⁸ 6 weeks before its co-administration with dolutegravir. However, a significant carry-over effect was unlikely as our participants were initially on efavirenz-based ART, which enhances piperazine metabolism by induction of CYP3A4¹⁹ and reduces piperazine's $t_{1/2}$ in pregnant women to a mean of 6 days.²⁰ Thirdly, this study was not designed to investigate

Table 1. Dolutegravir (DTG) exposure when administered alone (as DTG-based ART) compared with co-administration with dihydroartemisinin/piperazine (+ DP)

Pharmacokinetic parameter	GM (90% CI), N=12		GMR (90% CI) Sequence 3/Sequence 2	P value ^a
	DTG-based ART+ DP (Sequence 3)	DTG-based ART only (Sequence 2)		
AUC ₀₋₂₄ (ng·h/mL)	43 659 (39 939–47 726)	33,555 (29 479–38 196)	1.30 (1.11–1.52)	0.004
C _{max} (ng/mL)	3270 (3030–3529)	2504 (2213–2832)	1.31 (1.13–1.51)	0.001
C ₂₄ (ng/mL) ^b	701 (593–830)	495 (404–606)	1.42 (1.09–1.85)	0.008
T _{max} (h)	2.6 (2.1–3.3)	2.3 (1.8–3.1)	1.13 (0.80–1.61)	0.379
t _{1/2} (h)	9.9 (8.8–11.1)	9.0 (8.2–9.9)	1.10 (0.95–1.27)	0.063
CL/F (L/h)	1.15 (1.05–1.25)	1.49 (1.31–1.70)	0.77 (0.66–0.90)	0.004

GM, geometric mean. Bold represents statistical significance.

^aPaired *t*-test.

^bOne participant had an implausible DTG C₂₄ due to dosing prior to the 24 h sample. This value was imputed from the previous pre-dose concentration.

any potential impact of modestly increased dolutegravir exposure on long-term effects including maternal weight gain and metabolic syndrome.

The combination of dolutegravir-based ART and dihydroartemisinin/piperazine was well tolerated in this study, with no observed serious adverse events or changes in HIV viral load. Adverse events were mild to moderate and consistent with previously approved drug labels for both treatments.

In conclusion, dihydroartemisinin/piperazine can be administered with dolutegravir-based ART in PLWH, as the modest increase in dolutegravir exposure, similar to pharmacokinetic parameter values published previously, ensures its efficacy without any safety concerns observed in this small study.

Acknowledgements

We would like to thank all study participants for their willingness to take part in the study. We thank the Lighthouse Trust team in Zomba, Malawi (Dr Beatrice Mwangomba and Mr Thomas Chaweza) as well as the Malawi Department of Health's HIV Unit (Dr Thokozani Kalua) for their support during the implementation of the study. The Medical Dictionary for Regulatory Activities terminology (MedDRA[®]) used in this study is an international medical terminology developed under the auspices of the International Council for Harmonisation of Technical Requirements for Pharmaceuticals for Human Use (ICH). The MedDRA[®] trademark is registered by ICH.

Funding

This IMPROVE DDI study was funded by the EDCTP2 programme supported by the EU [Grant number TMA2017CDF-1897 (PENETRATE study grant) to C.G.B.]. The views and opinions of authors expressed herein do not necessarily state or reflect those of EDCTP. Further support for the dolutegravir assay was obtained from the Thrasher Research Fund through an Early Career Award (award number 15567) to C.G.B. This study piloted the Toolkit in Action and PK Guide developed by the WorldWide Antimalarial Resistance Network, co-funded by the ExxonMobil Foundation.

Transparency declarations

All authors declare no conflict of interest.

Supplementary data

Figures S1 and S2, Tables S1 to S3 and Text S1 and S2 are available as Supplementary data at JAC Online.

References

- Hochman S, Kim K. The impact of HIV and malaria coinfection: what is known and suggested venues for further study. *Interdiscip Perspect Infect Dis* 2009; **2009**: 617954.
- Laar AK, Grant FE, Addo Y et al. Predictors of fetal anemia and cord blood malaria parasitemia among newborns of HIV-positive mothers. *BMC Res Notes* 2013; **6**: 350.
- Hill J, Kuile FOT. Dihydroartemisinin-piperazine holds promise as an option for malaria prevention in pregnancy. *Evid Based Med* 2016; **21**: 146–7.
- ClinicalTrials.gov. Improving PRegnancy Outcomes With PReVEntive Therapy in Africa-2 (IMPROVE-2). <https://clinicaltrials.gov/ct2/show/NCT04158713>.
- González R, Nhampossa T, Mombo-Ngoma G et al. Evaluation of the safety and efficacy of dihydroartemisinin-piperazine for intermittent preventive treatment of malaria in HIV-infected pregnant women: protocol of a multicentre, two-arm, randomised, placebo-controlled, superiority clinical trial (MAMAH project). *BMJ Open* 2021; **11**: e053197.
- Tarning J, Ashley EA, Lindegardh N et al. Population pharmacokinetics of piperazine after two different treatment regimens with dihydroartemisinin-piperazine in patients with *Plasmodium falciparum* malaria in Thailand. *Antimicrob Agents Chemother* 2008; **52**: 1052–61.
- Walimbwa SI, Lamorde M, Waitt C et al. Drug interactions between dolutegravir and artemether-lumefantrine or artesunate-amodiaquine. *Antimicrob Agents Chemother* 2019; **63**: e01310-18.
- Van Lunzen J, Maggiolo F, Arribas JR et al. Once daily dolutegravir (S/GSK1349572) in combination therapy in antiretroviral-naïve adults with HIV: planned interim 48 week results from SPRING-1, a dose-ranging, randomised, phase 2b trial. *Lancet Infect Dis* 2012; **12**: 111–8.
- Deshpande S, Kuppast B. 4-Aminoquinolines: an overview of antimalarial chemotherapy. *Med Chem* 2016; **6**. <https://doi.org/10.4172/2161-0444.1000315>.
- FDA. Statistical Approaches to Establishing Bioequivalence. 2001. <https://www.fda.gov/regulatory-information/search-fda-guidance-documents/statistical-approaches-establishing-bioequivalence>.

- 11** Wen X, Wang J-S, Neuvonen PJ *et al*. Isoniazid is a mechanism-based inhibitor of cytochrome P450 1A2, 2A6, 2C19 and 3A4 isoforms in human liver microsomes. *Eur J Clin Pharmacol* 2002; **57**: 799–804.
- 12** Ribera E, Podzamczar D. Mechanism of action, pharmacology and interactions of dolutegravir. *Enferm Infecc Microbiol Clin* 2015; **33**: 2–8.
- 13** Elliot ER, Cerrone M, Else L *et al*. Pharmacokinetics of dolutegravir with and without darunavir/cobicistat in healthy volunteers. *J Antimicrob Chemother* 2019; **74**: 149–56.
- 14** Reese MJ, Savina PM, Generaux GT *et al*. In vitro investigations into the roles of drug transporters and metabolizing enzymes in the disposition and drug interactions of dolutegravir, a HIV integrase inhibitor. *Drug Metab Dispos* 2013; **41**: 353–61.
- 15** Senarathna SMDKG, Page-Sharp M, Crowe A. The interactions of P-glycoprotein with antimalarial drugs, including substrate affinity, inhibition and regulation. *PLoS One* 2016; **11**: e0152677.
- 16** Waitt C, Orrell C, Walimbwa S *et al*. Safety and pharmacokinetics of dolutegravir in pregnant mothers with HIV infection and their neonates: a randomised trial (DolPHIN-1 study). *PLoS Med* 2019; **16**: e1002895.
- 17** Mulligan N, Best BM, Wang J *et al*. Dolutegravir pharmacokinetics in pregnant and postpartum women living with HIV. *AIDS* 2018; **32**: 729–37.
- 18** Pan African Clinical Trials Registry (PACTR). IMPROVE DDI sub-study: dihydroartemisinin piperazine (DP)-dolutegravir-based ART drug-drug interactions in pregnancy (PACTR201910580840196). <https://pactr.samrc.ac.za/Search.aspx>.
- 19** Dooley KE, Flexner C, Andrade AS. Drug interactions involving combination antiretroviral therapy and other anti-infective agents: repercussions for resource-limited countries. *J Infect Dis* 2008; **198**: 948–61.
- 20** Kajubi R, Huang L, Jagannathan P *et al*. Antiretroviral therapy with efavirenz accentuates pregnancy-associated reduction of dihydroartemisinin-piperazine exposure during malaria chemoprevention. *Clin Pharmacol Ther* 2017; **102**: 520–8.