

Neither Branded Nor Generic Lopinavir/Ritonavir Produces Adequate Lopinavir Concentrations at a Reduced Dose of 200/50 mg Twice Daily

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Abstract: We assessed pharmacokinetic (PK) parameters of reduced dose lopinavir/ritonavir (LPV/r) and compared generic and branded tablets. Twenty HIV-infected patients using protease inhibitors with HIV RNA <50 copies per milliliter were randomized to generic or branded LPV/r 200/50mg twice daily (BID). At week 2, PK-sampling was performed. Patients crossed over to the other arm until week 12, with another PK-sampling at week 4. Subtherapeutic lopinavir concentrations were observed in 10/40 samples. PK parameters were comparable between branded and generic tablets. All patients remained virologically suppressed at week 12. In conclusion, LPV/r 200/50mg BID does not lead to adequate lopinavir plasma concentrations. Generic and branded LPV/r have comparable PK-parameters.

Key Words: clinical pharmacology, dose reduction, generics, HIV, Thailand

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INTRODUCTION

Lopinavir (LPV) is an HIV protease inhibitor (PI), coformulated with a subtherapeutic dose of ritonavir (RTV), another PI. RTV increases plasma concentrations of LPV by inhibiting its cytochrome P450 (CYP-450)-mediated metabolism.¹ This combination (LPV/r) has good efficacy and safety in treatment-naïve and treatment-experienced patients.^{2–4}

Dose reductions of several PIs in Thai HIV-1-infected patients result in adequate pharmacokinetic (PK) parameters due to lower body weight and different pharmacogenetic properties compared with Caucasian populations, with continued virological suppression, reduced toxicity, and lower cost.^{5–10}

The tablet formulation of LPV/r (200/50 mg) is bioequivalent to the soft-gel capsules (SGC), heat-stable, and its PK profile is not affected by food.¹¹ Despite significant price reductions for low-income and middle-income countries, the tablets are still expensive. Furthermore, due to a compulsory license, Abbott only markets the pediatric formulation (LPV/r tablets, 100/25 mg) in Thailand. The pharmaceutical company Matrix in India has developed a generic tablet formulation of LPV/r (200/50 mg). The use of these generic LPV/r tablets standard dose (400/100 mg twice daily) in Thai HIV-1-infected patients resulted in a median LPV minimum plasma concentration (C_{min}) of 7.2 mg/L,¹² well above the therapeutic concentration of 1.0 mg/L for PI-naïve patients and more than 10,000-fold higher than the protein binding-adjusted IC_{50} of wild-type HIV (0.69 ng/mL).^{13,14} We conducted the present study to determine whether lower dosing of generic LPV/r in Thai HIV-1-infected patients could achieve adequate concentrations and to assess the quality of the generic formulation.

METHODS

This was a single centre, open-label, prospective, 2-arm, randomized, cross-over PK study. The study was approved by

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the Ethics Committee of the Faculty of Medicine, Chulalongkorn University, Bangkok, Thailand. All participants provided written informed consent. The study was registered with ClinicalTrials.gov (NCT01159275). As this was a proof-of-concept study, we aimed to include 20 patients. Patients were screened at our clinic in Bangkok, Thailand. HIV-infected patients older than 18 years, using a PI-containing regimen for at least 4 weeks before study entry, without an AIDS-defining illness, and with plasma HIV RNA <50 copies per milliliter for at least 24 weeks before study entry were eligible. Study visits were at baseline, weeks 2, 4, and 12.

We collected demographic data and performed urinalysis and pregnancy testing. Participants were randomized at baseline to arm 1 or 2. In addition to a nucleoside reverse transcriptase inhibitor backbone, participants in arm 1 received generic LPV/r (200/50 mg) 1 tablet twice daily, and participants in arm 2 received branded LPV/r (Aluvia, LPV/r tablets, 100/25 mg, Abbott Laboratories, Abbott Park, IL), 2 tablets twice daily. For technical reasons, the participants were not blinded to the product received. After sample collection for PK analysis of LPV and RTV at week 2, patients crossed-over to the other study arm. At week 4, second PK analysis was performed. On both PK sample collection days, adherence was assessed by self-report, and adherence rates were calculated. Participants continued with the study regimen until week 12. After this, patients were switched back to their prestudy medication. Throughout the study, clinical and laboratory assessments (ie, liver transaminases, creatinine, hematology, CD4, and fasting blood glucose and lipids) were performed to evaluate the safety and tolerability of the study medication. Plasma HIV RNA was determined at week 12.

PK Analysis

PK analyses were performed at weeks 2 and 4. Patients were provided with standard meals throughout the day of sample collection, and the morning dose was taken under supervision of clinic staff. Six milliliter of blood was collected by venipuncture in a lithium-heparin or heparin-sodium tube shortly before administering the dose and at 1, 2, 3, 4, 6, 8, 10, and 12 hours after drug intake. Blood samples were centrifuged at 3000 revolutions per minute (rpm) at 20°C for 10 minutes. Plasma was transferred to a polypropylene tube and stored at -80°C until processing. The quantitative determination of LPV and RTV in plasma was carried out using a validated high-performance liquid chromatography method.¹⁵ The LPV concentration was linear over the range of 0.1–30.0 mg/L. The lower limit of quantification for LPV was 0.1 mg/L. The HIV Netherlands Australia Thailand Research Collaboration laboratory participates in an international quality control and quality assessment program and has been cross-validated with other PK laboratories.¹⁶ The following PK parameters were determined by noncompartmental analysis using Stata version 11.0 (StataCorp LP, College Station, TX): area under the plasma concentration–time curve from 0 to 12 hours (AUC_{0-12}), the maximum plasma drug concentration (C_{max}), the trough plasma drug concentration (C_{trough}), the time to reach maximum plasma drug concentration (T_{max}), and apparent elimination half life ($T_{1/2}$).

Statistical Analysis

Statistical analyses were performed using Stata version 11.0. Median [interquartile range (IQR)] and frequency (%) were used to describe the demographic characteristics for continuous and categorical data, respectively. Analyses of variance (ANOVA) were performed on the log (natural)-transformed PK parameters AUC_{0-12} , C_{max} , and C_{trough} . The analyses of variance model included sequence, formulation, and period as fixed effects and subjects within sequence as random effect. To detect statistically significant differences among different doses, the Kruskal–Wallis test was used. Geometric mean ratio (GMR) and 90% confidence interval (CI) for AUC_{0-12} , C_{max} , and C_{trough} for LPV and RTV were calculated. PK parameters of both products was compared using bioequivalence analysis; the products were considered comparable when the 90% CI of the GMR of LPV AUC_{0-12} and C_{max} of the generic tablet relative to the branded tablet were within the range of 0.80 to 1.25.¹⁷

RESULTS

Demographic and Clinical Characteristics

Twenty patients (8 women and 12 men) were included and randomized to one of the study arms (10 to arm 1 and 10 to arm 2). Median (IQR) age of participants was 38.6 (34.4–47.5) years, median (IQR) weight was 59.8 (52.9–62.0) kg, and median (IQR) CD4 count was 578 (476–795) cells per cubic millimeter. All patients had HIV RNA <50 copies per milliliter. Before study entry, 17 patients were using LPV/r (400/100 mg twice daily) for a median (IQR) of 18 (14–47) months; 3 patients were using saquinavir (SQV)/r for a median (IQR) of 30 (29–31) months, all with a nucleoside reverse transcriptase inhibitor backbone. No patients were lost to follow-up, and none discontinued the medication during the study period.

PK Results

PK parameters for generic and branded LPV/r are shown in Table 1. Generic and branded tablets showed comparable PK parameters. Mean (SD) C_{trough} was 1.5 (0.6) mg/L for generic tablets and 1.6 (0.9) mg/L for branded tablets ($P = 0.92$).

Subtherapeutic plasma concentrations of LPV (<1.0 mg/L) were detected in 8 patients; 2 patients although using generic tablets and 4 patients although using branded tablets. Two other patients had subtherapeutic concentrations on both, resulting in a total of 10 of 40 samples (25%). The lowest LPV plasma concentration measured was 0.25 mg/L. At the time of measured subtherapeutic concentration, all 8 patients reported adherence rates of >90%, and 7 of 8 patients reported adherence rates of 100%.

Results of the bioequivalence analysis comparing branded and generic tablets are shown in Table 2. The 90% CI of the GMR for generic LPV AUC_{0-12} , C_{max} , and C_{min} were 0.92–1.09, 0.90–1.07, and 0.76–1.31, respectively.

Using data from a previous study by our group, we compared PK parameters of different doses of LPV/r (data not shown).¹⁰ Compared with LPV/r SGC 400/100 mg twice daily, LPV/r SGC 266/66 mg twice daily resulted in a 44.1% decreased

TABLE 1. PK Parameters of Generic and Branded LPV/r 200/50 mg Twice Daily

PK Parameter	Generic Tablets Mean (SD)	Branded Tablets Mean (SD)	P*
LPV			
AUC ₀₋₁₂ (mg·h·L ⁻¹)	46.6 (10.7)	45.1 (16.9)	0.98
CV (%)	23.0	37.5	
C _{max} (mg/L)	6.2 (1.4)	6.1 (2.2)	0.91
CV (%)	22.6	36.1	
C _{trough} (mg/L)	1.5 (0.6)	1.6 (0.9)	0.92
CV (%)	40.0	56.3	
T _{1/2} (hr)	3.6 (0.9)	3.7 (1.2)	0.52
CV (%)	25.0	32.4	
RTV			
AUC ₀₋₁₂ (mg·h·L ⁻¹)	1.98 (0.5)	1.9 (0.7)	0.77
CV (%)	25.3	36.3	
C _{max} (mg/L)	0.28 (0.08)	0.26 (0.1)	0.46
CV (%)	28.6	38.5	
C _{trough} (mg/L)	0.07 (0.02)	0.07 (0.03)	0.34
CV (%)	28.6	42.9	
T _{1/2} (hr)	4.8 (2.0)	4.2 (1.3)	0.21
CV (%)	41.7	31.0	

*The P value evaluated by analysis of variance. T_{1/2}, the elimination half-life; CV: coefficient of variation.

LPV AUC₀₋₁₂, a 36.0% decreased LPV C_{max} and a 49.1% decreased LPV C_{trough}. LPV/r generic tablets 200/50 mg twice daily resulted in a 63.5% decreased AUC₀₋₁₂, a 56.6% decreased LPV C_{max} and a 70.2% decreased LPV C_{trough} compared with LPV/r SGC 400/100 mg twice daily.

Virological Response, Safety, and Tolerability

At week 12, all patients had plasma HIV-1 RNA <50 copies per milliliter. One patient experienced an adverse event (jaundice and elevated liver transaminases) during the study, which was unrelated to study drugs and caused by acute hepatitis C virus infection. Median (IQR) triglycerides levels decreased significantly from baseline [175 (131–285) mg/dL] to week 12 [115 (84–217) mg/dL], P < 0.001. Decreases in total cholesterol and low-density lipoprotein cholesterol were not significant.

DISCUSSION

In this study, we observed inadequate plasma concentrations with LPV/r 200/50 mg twice daily, irrespective of the formulation used, and demonstrated that generic and branded LPV/r tablets have comparable PK parameters.

Using standard dose of generic tablets of LPV/r, we have previously shown encouraging PK parameters and short-term efficacy.¹² Reduced dose of LPV/r in Thai HIV-1-infected patient has been described as well in a PK study of SGC LPV/r 400/100 or 266/66 mg twice daily coadministered with SQV 1000 or 600 mg twice daily.¹⁰ The number of patients with adequate LPV concentrations was similar in both arms, irrespective of LPV or SQV dose received.

Thai HIV-1-infected patients taking recommended doses of several PIs show higher plasma concentrations, and dose reduction of these PIs result in maintained viral efficacy.⁶⁻⁸ In these studies, the doses of SQV, atazanavir, and indinavir were reduced although maintaining the RTV dose. Compared with PIs such as SQV and darunavir, LPV plasma concentrations are to a greater extent dependent on the RTV dose used, that is, a higher dose of RTV will lead to a higher LPV plasma concentration.¹⁸ In our study, we reduced both the LPV and RTV doses, which could partly explain the found subtherapeutic LPV concentrations.

Based on currently available data on the association between LPV concentration and virological efficacy in patients with HIV subtype B, current guidelines recommend a minimum LPV trough concentration of 1.0 mg/L for PI-naive patients.¹³ Due to the small sample size and limited follow-up time of our study, we cannot draw a definite conclusion about the virological implication of the found subtherapeutic concentrations. Although the lowest measured LPV concentration in our study (0.25 mg/L) was 355-fold higher than the IC₅₀ of wild-type virus,¹⁴ prolonged exposure to poor LPV plasma concentrations results in a higher risk of virological failure.¹⁹⁻²² Furthermore, the susceptibility of the CRF01_AE virus (the main HIV subtype found in Thailand) to LPV might be reduced compared with that of subtype B,²³ although a difference in virological response rate to LPV according to HIV subtype in a clinical setting has not been established.²⁴

As all patients with subtherapeutic levels reported good adherence, we can consider the possibility of nonadherence as a cause for subtherapeutic concentrations as less likely. However,

TABLE 2. Relative Bioavailability and 90% Confidence Interval for the Geometric Mean Ratio for LPV and RTV

PK Parameters	Geometric Mean Ratio		Relative Bioavailability	
	Generic Tablets	Branded Tablets	Point Estimate	90% Confidence Interval
LPV				
Log10 AUC ₀₋₁₂	1.64	1.64	1.00	0.92 to 1.09
Log10 C _{max}	0.79	0.75	1.01	0.90 to 1.07
Log10 C _{trough}	0.19	0.22	0.87	0.76 to 1.31
RTV				
Log10 AUC ₀₋₁₂	0.25	0.23	1.07	0.93 to 1.18
Log10 C _{max}	2.41	2.36	1.02	0.93 to 1.18
Log10 C _{trough}	1.85	1.80	1.03	0.88 to 1.13

as we did not perform pill count, socially desirable responding cannot be excluded. Extraordinarily high levels of adherence are required using the 200/50 mg twice daily dose, and this dose would be substantially less forgiving of nonadherence compared with the standard dose.

PK profiles of alternative dosages for LPV/r have been investigated in other populations as well. One of these describes the PK profile of LPV/r in 16 antiretroviral naive patients receiving LPV/r 200/100 mg twice daily. After 3 weeks, the mean (\pm SD) LPV C_{trough} was 3.74 ± 3.46 mg/L, and all patients achieved virological suppression (plasma HIV RNA below 50 copies/mL) after 48 weeks of treatment,³ suggesting 200 mg LPV twice daily is sufficient, if there is a sufficient dose of RTV for boosting of LPV concentrations.

In a study in HIV-uninfected volunteers, using 200/50 mg twice daily, the C_{trough} was 70% lower compared with 400/100 mg twice daily, supporting that adequate RTV concentrations are pivotal when reducing the LPV dose.²⁵ Using LPV/r 200/150 mg twice daily, a 26% reduction of LPV plasma concentrations was observed compared with 400/100 mg twice daily, due to higher RTV concentrations. All patients using this dosage had LPV plasma concentrations >1.0 mg/L. This study was not powered to assess the different rates of adverse events between the different doses.

Although we did not compare generic and branded products using the approved dose of LPV/r, that is, 400/100 mg twice daily, our bioequivalence analysis demonstrated that the generic and branded tablets result in comparable PK parameters. These data are particularly important for clinicians working in settings where the branded tablets are not available due to compulsory licensing or cost. The availability of safe and effective generic alternatives to branded second-line treatment will play an important role in the scaling-up of second-line treatment in low-income and middle-income countries.

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REFERENCES

- Sham HL, Kempf DJ, Molla A, et al. ABT-378, a highly potent inhibitor of the human immunodeficiency virus protease. *Antimicrob Agents Chemother.* 1998;42:3218–3224.
- Benson CA, Deeks SG, Brun SC, et al. Safety and antiviral activity at 48 weeks of lopinavir/ritonavir plus nevirapine and 2 nucleoside reverse-transcriptase inhibitors in human immunodeficiency virus type 1-infected protease inhibitor-experienced patients. *J Infect Dis.* 2002;185:599–607.
- Murphy RL, Brun S, Hicks C, et al. ABT-378/ritonavir plus stavudine and lamivudine for the treatment of antiretroviral-naive adults with HIV-1 infection: 48-week results. *AIDS.* 2001;15:F1–F9.
- Walmsley S, Bernstein B, King M, et al. Lopinavir-ritonavir versus nelfinavir for the initial treatment of HIV infection. *N Engl J Med.* 2002;346:2039–2046.
- Ananworanich J, Gayet-Ageron A, Ruxrungtham K, et al. Long-term efficacy and safety of first-line therapy with once-daily saquinavir/ritonavir. *Antivir Ther.* 2008;13:375–380.
- Autar RS, Boffito M, Hassink E, et al. Interindividual variability of once-daily ritonavir boosted saquinavir pharmacokinetics in Thai and UK patients. *J Antimicrob Chemother.* 2005;56:908–913.
- Avihingsanon A, van der Lugt J, Kerr SJ, et al. A low dose of ritonavir-boosted atazanavir provides adequate pharmacokinetic parameters in HIV-1-infected Thai adults. *Clin Pharmacol Ther.* 2009;85:402–408.
- Boyd M, Mootsikapun P, Burger D, et al. Pharmacokinetics of reduced-dose indinavir/ritonavir 400/100 mg twice daily in HIV-1-infected Thai patients. *Antivir Ther.* 2005;10:301–307.
- Hartkoorn RC, Kwan WS, Shallcross V, et al. HIV protease inhibitors are substrates for OATP1A2, OATP1B1 and OATP1B3 and lopinavir plasma concentrations are influenced by SLCO1B1 polymorphisms. *Pharmacogenet Genomics.* 2010;20:112–120.
- van der Lugt J, Autar RS, Ubolyam S, et al. Pharmacokinetics and short-term efficacy of a double-boosted protease inhibitor regimen in treatment-naive HIV-1-infected adults. *J Antimicrob Chemother.* 2008;61:1145–1153.
- Klein CE, Chiu YL, Awni W, et al. The tablet formulation of lopinavir/ritonavir provides similar bioavailability to the soft-gelatin capsule formulation with less pharmacokinetic variability and diminished food effect. *J Acquir Immune Defic Syndr.* 2007;44:401–410.
- van der Lugt J, Lange J, Avihingsanon A, et al. Plasma concentrations of generic lopinavir/ritonavir in HIV type-1-infected individuals. *Antivir Ther.* 2009;14:1001–1004.
- Panel on Antiretroviral Guidelines for Adults and Adolescents. Guidelines for the Use of Antiretroviral Agents in HIV-1-Infected Adults and Adolescents. Bethesda, MD: Department of Health and Human Services. Updated January 10, 2011. Available at: <http://aidsinfo.nih.gov/contentfiles/AdultandAdolescentGL.pdf>. Accessed January 31, 2011.
- Hickman D, Vasavanonda S, Nequist G, et al. Estimation of serum-free 50-percent inhibitory concentrations for human immunodeficiency virus protease inhibitors lopinavir and ritonavir. *Antimicrob Agents Chemother.* 2004;48:2911–2917.
- Droste JA, Verweij-Van Wissen CP, Burger DM. Simultaneous determination of the HIV drugs indinavir, amprenavir, saquinavir, ritonavir, lopinavir, nelfinavir, the nelfinavir hydroxymetabolite M8, and nevirapine in human plasma by reversed-phase high-performance liquid chromatography. *Ther Drug Monit.* 2003;25:393–399.
- Burger D, Teulen M, Eerland J, et al. The International Interlaboratory Quality Control Program for Measurement of Antiretroviral Drugs in Plasma: a global proficiency testing program. *Ther Drug Monit.* 2011;33:239–243.
- US Food and Drug Administration. Bioavailability and bioequivalence studies for orally administered drug products—general considerations Available at: <http://www.fda.gov/downloads/Drugs/GuidanceComplianceRegulatoryInformation/Guidances/ucm070124.pdf>. Accessed September 5, 2011.
- Hill A, van der Lugt J, Sawyer W, et al. How much ritonavir is needed to boost protease inhibitors? Systematic review of 17 dose-ranging pharmacokinetic trials. *AIDS.* 2009;23:2237–2245.
- Breilh D, Pellegrin I, Rouzes A, et al. Virological, intracellular and plasma pharmacological parameters predicting response to lopinavir/ritonavir (KALEPHAR study). *AIDS.* 2004;18:1305–1310.
- Fabbiani M, Di Giambenedetto S, Bracciale L, et al. Pharmacokinetic variability of antiretroviral drugs and correlation with virological outcome: 2 years of experience in routine clinical practice. *J Antimicrob Chemother.* 2009;64:109–117.
- Gonzalez de Requena D, Gallego O, Valer L, et al. Prediction of virological response to lopinavir/ritonavir using the genotypic inhibitory quotient. *AIDS Res Hum Retroviruses.* 2004;20:275–278.
- van Zyl GU, van Mens TE, McIlleron H, et al. Low lopinavir plasma or hair concentrations explain second-line protease inhibitor failures in a resource-limited setting. *J Acquir Immune Defic Syndr.* 2011;56:333–339.
- Jinnopat P, Isarangkura-na-ayuthaya P, Utachee P, et al. Impact of amino acid variations in Gag and protease of HIV type 1 CRF01_AE strains on drug susceptibility of virus to protease inhibitors. *J Acquir Immune Defic Syndr.* 2009;52:320–328.
- Dierynck I, De Meyer S, Lathouwers E, et al. In vitro susceptibility and virological outcome to darunavir and lopinavir are independent of HIV type-1 subtype in treatment-naive patients. *Antivir Ther.* 2010;15:1161–1169.
- Jackson A, Hill A, Puls R, et al. Pharmacokinetics of plasma lopinavir/ritonavir following the administration of 400/100 mg, 200/150 mg and 200/50 mg twice daily in HIV-negative volunteers. *J Antimicrob Chemother.* 2010;66:635–640.