Pharmacokinetics of Adjusted-Dose Lopinavir-Ritonavir Combined with Rifampin in Healthy Volunteers


Updated information and services can be found at:
http://aac.asm.org/content/48/5/1553

REFERENCES

These include:
This article cites 17 articles, 4 of which can be accessed free at:
http://aac.asm.org/content/48/5/1553#ref-list-1

CONTENT ALERTS

Receive: RSS Feeds, eTOCs, free email alerts (when new articles cite this article), more»

Information about commercial reprint orders: http://journals.asm.org/site/misc/reprints.xhtml
To subscribe to another ASM Journal go to: http://journals.asm.org/site/subscriptions/
Pharmacokinetics of Adjusted-Dose Lopinavir-Ritonavir Combined with Rifampin in Healthy Volunteers

C. J. L. la Porte,1,2* E. P. H. Colbers,3 R. Bertz,4 D. S. Voncken,3 K. Wikstrom,4 M. J. Boeree,2,5 P. P. Koopmans,2,6 Y. A. Hekster,1,2 and D. M. Burger1,2

Department of Clinical Pharmacy1 and Department of General Medicine,6 University Medical Centre Nijmegen, Nijmegen University Centre for Infectious Diseases,2 Farma Research BV,3 and University Lung Centre Dekkerswald,5 Nijmegen, The Netherlands, and Department of Clinical Pharmacokinetics and Antiviral Global Development, Abbott Laboratories, Abbott Park, Illinois

Received 8 October 2003/Returned for modification 4 December 2003/Accepted 3 February 2004

Coadministration of lopinavir-ritonavir, an antiretroviral protease inhibitor, at the standard dose (400/100 mg twice a day [BID]) with the antituberculous agent rifampin is contraindicated because of a significant pharmacokinetic interaction due to induction of cytochrome P450 3A by rifampin. In the present study, two adjusted-dose regimens of lopinavir-ritonavir were tested in combination with rifampin. Thirty-two healthy subjects participated in a randomized, two-arm, open-label, multiple-dose, within-subject controlled study. All subjects were treated with lopinavir-ritonavir at 400/100 mg BID from days 1 to 15. From days 16 to 24, the subjects in arm 1 received lopinavir-ritonavir at 800/200 mg BID in a dose titration, and the subjects in arm 2 received lopinavir-ritonavir at 400/400 mg BID in a dose titration. Rifampin was given at 600 mg once daily to all subjects from days 11 to 24. The multiple-dose pharmacokinetics of lopinavir, ritonavir, and rifampin were assessed. Twelve of 32 subjects withdrew from the study. For nine subjects lopinavir-ritonavir combined with rifampin resulted in liver enzyme level elevations. Pharmacokinetic data for 19 subjects were evaluable.

The treatment of human immunodeficiency virus (HIV)-infected individuals has improved greatly over the past several years. With the development of antiretroviral agents in different classes, more options for the effective suppression of the virus have become available (6). However, many problems remain to be solved. One of them is the treatment of patients presenting with HIV infection and coinfections. Tuberculosis is a significant opportunistic infection in HIV-infected individuals in developing countries and, to a lesser extent, in developed countries (1, 3, 11, 15). For public health reasons, active tuberculosis must be treated immediately (3). The treatment of HIV infection at a standard dosage of 400/100 mg twice a day (BID) with the antituberculous agent rifampin is contraindicated because of a significant pharmacokinetic interaction due to induction of cytochrome P450 3A by rifampin. In the present study, two adjusted-dose regimens of lopinavir-ritonavir were tested in combination with rifampin. Thirty-two healthy subjects participated in a randomized, two-arm, open-label, multiple-dose, within-subject controlled study. All subjects were treated with lopinavir-ritonavir at 400/100 mg BID from days 1 to 15. From days 16 to 24, the subjects in arm 1 received lopinavir-ritonavir at 800/200 mg BID in a dose titration, and the subjects in arm 2 received lopinavir-ritonavir at 400/400 mg BID in a dose titration. Rifampin was given at 600 mg once daily to all subjects from days 11 to 24. The multiple-dose pharmacokinetics of lopinavir, ritonavir, and rifampin were assessed. Twelve of 32 subjects withdrew from the study. For nine subjects lopinavir-ritonavir combined with rifampin resulted in liver enzyme level elevations. Pharmacokinetic data for 19 subjects were evaluable.

The treatment of human immunodeficiency virus (HIV)-infected individuals has improved greatly over the past several years. With the development of antiretroviral agents in different classes, more options for the effective suppression of the virus have become available (6). However, many problems remain to be solved. One of them is the treatment of patients presenting with HIV infection and coinfections. Tuberculosis is a significant opportunistic infection in HIV-infected individuals in developing countries and, to a lesser extent, in developed countries (1, 3, 11, 15). For public health reasons, active tuberculosis must be treated immediately (3). The treatment of HIV infection can be postponed on the basis of CD4 cell counts and the viral load. However, depending on the clinical and biochemical parameters for coinfected patients, simultaneous treatment of both infections can become indicated in particular situations. The combination of antiretroviral therapy with therapy against tuberculosis is complex. In particular, the use of rifampin is hampered due to significant drug-drug interactions. Rifampin is a first-line antibacterial agent for the treatment of tuberculosis and acts by inhibiting the DNA-dependent RNA polymerase of the microorganism (2). Rifampin is a strong inducer of cytochrome P450 (CYP)-mediated metabolism of other agents; in particular, the CYP3A isoenzyme is subject to induction. Rifampin metabolism itself is not dependent on CYP3A; nevertheless, autoinduction of cholinesterase- and B esterase-mediated metabolism of rifampin has been shown (2). Because of its CYP3A-inducing effects, rifampin is known to produce significant pharmacokinetic interactions with HIV protease inhibitors (PIs) and nonnucleoside reverse transcriptase inhibitors (NNRTIs) (13). These pharmacokinetic interactions may lead to subtherapeutic levels of these antiretroviral agents in plasma, and for this reason, rifampin in combination with most PIs and NNRTIs is contraindicated. This clearly limits the options for highly active antiretroviral therapy in Mycobacterium tuberculosis-coinfected HIV-infected patients. Data on the adjusted dosages that generally result in therapeutic levels of the antiretrovirals in plasma have been published for the PI saquinavir (17) and the NNRTI efavirenz (10). Pharmacokinetic interactions between rifampin and nucleoside reverse transcriptase inhibitors (NRTIs) are less pronounced, since these agents do not undergo appreciable oxidative metabolism (4, 14). A study of the interaction of rifampin with T-20 (enfuvirtide) did not reveal clinically significant changes in the pharmacokinetics of T-20 (Fuzeon; Summary of Product Characteristics, 2003; Roche Registration Limited, Welwyn Garden City, United Kingdom).

Lopinavir-ritonavir is a formulation of two PIs approved for the treatment of HIV infection at a standard dosage of 400/100
mg twice a day (BID) in combination with other antiretrovirals. The coformulation of lopinavir-ritonavir is available as capsules with a dose of 133 mg of lopinavir and 33 mg of ritonavir per capsule. Lopinavir is mainly dependent on CYP3A for its metabolism, and ritonavir is a strong inhibitor of CYP3A. For this reason, ritonavir is coformulated with lopinavir (Kaletra Product Information, 2000; Abbott Laboratories, Chicago, Ill.), resulting in sustained and elevated plasma lopinavir levels. The pharmacokinetic interaction between rifampin and lopinavir-ritonavir has been studied previously (R. Bertz, A. Hsu, W. Lam, L. Williams, C. Renz, M. D. Karol, S. Dutta, A. Carr, Y. Zhang, Q. Wang, S. Schweitzer, C. Foit, A. Andre, B. Bernstein, G. Granneman, and E. Sun, 5th Int. Congr. Drug Ther. HIV Infect., abstr. P291, 2000). The area under the concentration-time curve (AUC) and the minimum concentration in plasma ($C_{\text{min}}$) for lopinavir in healthy subjects were reduced 75% and 99%, respectively, as a result of coadministration of rifampin at 600 mg once daily (QD) with lopinavir-ritonavir at 400/100 mg BID. This is the direct result of the strong induction of CYP3A by rifampin, which overcomes the inhibition of CYP3A by low-dose ritonavir. Therefore, rifampin combined with lopinavir-ritonavir in the standard-dose regimen is contraindicated. The objective of the present study was to investigate in healthy subjects the pharmacokinetics of two adjusted-dose regimens of lopinavir-ritonavir in combination with rifampin in comparison to the standard dose of lopinavir-ritonavir without rifampin.

### MATERIALS AND METHODS

**Study design.** The study was designed as a randomized, phase I, open-label, two-arm, within-subject controlled study with 32 healthy subjects, both males and females. See Table 1 for details of the study design. The study consisted of a run-in period of 10 days in which lopinavir-ritonavir at the standard dose (400/100 mg BID as three coformulated capsules) was given to all subjects. On study day 10, the steady-state pharmacokinetics of lopinavir and ritonavir were determined over the daytime 12-h dosing interval. During study days 11 to 15, the subjects continued fasting (no food or beverages) until 5 h after drug intake, at which time the subjects received a standardized lunch. The subjects received a standardized snack at 9 h following drug ingestion, and dinner was served after the last blood draw. Beverages (i.e., water, orange juice, apple juice, coffee, tea, and milk) were allowed ad libitum from 5 h after dosing until the end of the confinement period.

**Selection of subjects.** This study was performed with healthy subjects. The inclusion criteria were the ability to sign voluntary informed consent; age 18 years or older; good health (i.e., the subject was not suffering from an acute or chronic illness and was not using medications); and a body mass index (BMI) lower than 30.0 for men and lower than 28.6 for women (body mass index is equal to weight [in kilograms]/height$^2$ [in square meters]). Female subjects could not become pregnant or breastfeed. Exclusion criteria were as follows: known hypersensitivity to lopinavir, ritonavir, or rifampin; positive test result for HIV; hepatitis B or C virus; a tuberculin skin test reaction of more than 15 mm or a history of tuberculosis; pregnancy or breastfeeding; body weight $\leq 50$ kg; use of contact lenses; a history of pancreatitis; a history of alcohol abuse; and one or more of the following laboratory test results: hemoglobin concentration, $\leq 10.0$ g/dl; leukocyte count, $\leq 9.0$ X10$^3$/liter; alanine aminotransferase (ALT) $\geq 3$ X the ULN; total bilirubin levels more than two times the ULN; alkaline phosphatase levels more than two times the ULN; serum creatinine levels more than 1.5 times the ULN; lipase levels more than two times the ULN; or total bilirubin levels more than two times the ULN.

**Blood sampling procedure.** For determination of lopinavir and ritonavir concentrations, blood samples (5 ml) were collected predosing and at 2, 4, 6, 8, 10, and 12 h postdosing on days 10 and 24. Rifampin concentrations were determined with the same samples used for determination of lopinavir and ritonavir concentrations. Blood samples (5 ml) were collected predosing and at 2, 4, 6, 8, 10, and 12 h postdosing on days 10 and 24. Rifampin concentrations were determined with the same samples used for determination of lopinavir and ritonavir concentrations on day 24. However, additional blood samples (5 ml) for the more precise determination of the maximum concentration in plasma ($C_{\text{max}}$ for rifampin) were drawn at 1 and 3 h postdosing on day 24. Subjects returned to the study location on study days 1, 3, 7, 13, 16, 17, 18, 20, and 22 for predose blood sampling (5 ml) for determination of lopinavir and ritonavir concentrations. Blood samples were centrifuged at approximately 1,000 X g for 10 min at 4°C. Plasma for determination of lopinavir and ritonavir concentrations was transferred to a labeled polypropylene tube and stored at $\approx-18^\circ$C within 2 h after collection. Plasma for determination of rifampin concentrations was transferred to a labeled polypropylene tube containing ascorbic acid and stored at $\approx-80^\circ$C within 2 h after collection.

### Table 1. Study design

<table>
<thead>
<tr>
<th>Randomisation</th>
<th>Regimen(s) administered on the following days:</th>
</tr>
</thead>
<tbody>
<tr>
<td>Arm 1</td>
<td>LPV/r, 533/133 mg BID; RIF, 600 mg QD</td>
</tr>
<tr>
<td></td>
<td>LPV/r, 400/200 mg BID; RIF, 600 mg QD</td>
</tr>
<tr>
<td></td>
<td>LPV/r, 667/167 mg BID; RIF, 600 mg QD</td>
</tr>
<tr>
<td></td>
<td>LPV/r, 800/200 mg BID; RIF, 600 mg QD</td>
</tr>
<tr>
<td>Arm 2</td>
<td>LPV/r, 400/200 mg BID; RIF, 600 mg QD</td>
</tr>
<tr>
<td></td>
<td>LPV/r, 400/400 mg BID; RIF, 600 mg QD</td>
</tr>
</tbody>
</table>

\*a From days 1 to 10, all subjects received lopinavir-ritonavir at 400/100 mg BID; and from days 11 to 15, all subjects received lopinavir-ritonavir at 400/100 mg BID plus rifampin at 600 mg QD. The subjects were randomized to arm 1 or arm 2 after day 15. On days 10 and 24, blood sampling was performed up to 12 post ingestion. Lopinavir-ritonavir (LPV/r) was coformulated in capsules, each of which contained 133 mg of lopinavir and 33 mg of ritonavir. A lopinavir-ritonavir dose of 400/100 mg BID therefore consisted of three capsules. Rifampin (RIF) was dosed as two capsules, each of which containing 300 mg of rifampin. \*b In the combinations with lopinavir-ritonavir at 400/200 mg BID, 400/300 mg BID, and 400/400 mg BID, the extra ritonavir was dosed as 100-mg ritonavir capsules in addition to the normal combination of lopinavir-ritonavir at 400/100 mg.
Bioanalysis. Plasma lopinavir and ritonavir levels were determined by a validated high-performance liquid chromatography (HPLC) method, which was a modified version of a previously published method (9). The modification consisted of a switch of the UV detection wavelength from 245 to 215 nm at 15.5 min, with a retention time of 14.4 min for ritonavir, and from 9.2 to 100% for lopinavir. The concentration of each agent could be measured without interference from the other drug. The lopinavir and ritonavir calibration curves were linear over a range of 0.045 to 30.0 mg/liter. The lower limit of quantification was 0.04 mg/liter for both lopinavir and ritonavir. Rates of recovery after extraction from plasma were 95% for lopinavir and 94% for ritonavir. The accuracies ranged from 99 to 101% for lopinavir, and from 92 to 100% for ritonavir, and the intra- and inter-day precisions ranged from 0.92 to 5.16% for lopinavir and from 1.51 to 5.14% for ritonavir. The interday precisions ranged from 0.1 to 1.57% for lopinavir and from 0 to 5.00% for ritonavir.

Plasma rifampin levels were measured by a validated HPLC method that was developed in the University Medical Centre Nijmegen but that has not yet been published. The method consisted of protein precipitation followed by reversed-phase HPLC with UV detection. Two hundred microliters of acetonitrile was added to 200 μl of plasma to precipitate protein. This mixture was vortexed for 20 s, and afterwards the mixture was centrifuged for 5 min. Fifty microliters of the clear supernatant was used for injection. Chromatographic analysis was performed on an Inertisil 5 ODS 2 analytical column (250 by 4.6 mm [inner diameter]; Varian, Bergens op;om, The Netherlands) protected with a Chromguard HPLC column (10 by 3 mm[inner diameter]; Varian). The mobile phase was a mixture of 0.1 M potassium dihydrogen phosphate (62%) and acetonitrile (38%). The flow rate was 1 ml/min, and the wavelength for UV detection was 334 nm. The rifampin retention time was 7.3 min. The rifampin calibration curve was linear over a range of 0.50 to 30.0 mg/liter. The lower limit of quantification for rifampin was 0.50 mg/liter. Recovery after extraction from plasma was 108.5%. Accuracy ranged from 101.3 to 102.2%, and intraday and interday precisions ranged from 2.84 to 3.65% and from 1.59 to 3.68%, respectively.

Safety monitoring and laboratory safety. The medical history, vital signs, a physical examination, and an electrocardiogram for each subject were obtained at screening. Laboratory tests were done at screening and all study visits (days 1, 3, 7, 10, 13, 16, 17, 18, 20, 22, and 24). Laboratory tests included tests for sodium, potassium, creatinine, total bilirubin, cholesterol, triglycerides, glucose, alkaline phosphatase, ASAT, ALAT, GGT, and amylase (pancreatic) levels; a whole-blood cell count; and urinalysis.

Additionally, subjects were asked about the occurrence of adverse events at each visit. Adverse events were assessed for intensity, according to the AIDS Clinical Trials Group classifications mild (symptoms do not interfere with daily activities), moderate (symptoms interfere with daily activities), and severe (symptoms markedly interrupt daily activities), and seriousness. Serious adverse events were defined as any untoward medical occurrence that at any dose resulted in death, that was life threatening, that required in-patient hospitalization or prolongation of existing hospitalization, that resulted in a persistent or significant disability or incapacity, or that was a congenital anomaly or birth defect. During the study, the occurrence of grade 2 toxicity, according to World Health Organization scales, would result in discontinuation of a subject from the study. For cholesterol and triglycerides, grade 3 toxicity was a reason to discontinue study medication.

Pharmacokinetic analysis. Values for the pharmacokinetic parameters of lopinavir, ritonavir, and rifampin were estimated by noncompartmental methods. The Cmax and the time to Cmax (Tmax) were determined directly from the plasma concentration-time data. Cmin and the morning predose observed trough concentration in plasma (C0) were also determined directly from the plasma concentration-time data. The area under the plasma concentration-time curve from time zero to 12 h postdosing (AUC(t2)) was calculated by use of the linear trapezoidal rule. The value of the peak-to-trough rate constant (β) was obtained from the slope of the least-squares regression of the logarithms of the plasma concentration-versus-time data for the 12-h interval, which was then used to calculate the half-life (τ1/2). The dosing interval or peak-to-trough τ1/2 was calculated as ln 2/β. The apparent oral clearance value (CL/F), where F is the bioavailability, was calculated by dividing the administered dose in a dosing interval by AUC(t2). CL/F was normalized for body weight (CL/F/kg) by dividing by the weight (in kilograms). The apparent volume of distribution (V/F) was calculated by dividing CL/F by β. V/F was normalized for body weight (V/F/kg) by dividing by the weight (in kilograms).

Statistical analysis. The pharmacokinetic data for lopinavir, ritonavir, and rifampin are presented as arithmetic means ± standard deviations and geometric means ± standard deviations. Data were logarithmically transformed for the calculation of geometric means. The median and interquartile ranges are presented for Cmax. The change in a pharmacokinetic variable for lopinavir or ritonavir from the administered value of lopinavir-ritonavir alone to the administration of the combination regimen with rifampin was analyzed by a paired t test for each of the study arms. A Wilcoxon signed-ranks test was used for Tmax. P values less than 0.05 were considered statistically significant. Variables included logarithmically transformed AUC(t2), Cmax, and nontransformed T1/2 and T1/2. The bioavailability ratio for the combination regimen relative to that for lopinavir alone was assessed by the two one-sided-tests procedure with 90% confidence intervals for AUC(t2). Cmax, and Cmax. For this purpose geometric mean ratios were calculated by dividing the geometric mean values for study day 24 by the geometric mean values for study day 10. The 90% confidence intervals of the geometric mean ratios were obtained by exponentiating the confidence limits for the differences in logarithmic means. The geometric mean ratios together with the 90% confidence intervals were compared to the range of 0.80 to 1.25 to determine whether the lopinavir-ritonavir dose regimens combined with rifampin met the criteria for bioequivalence to the standard clinical dose of lopinavir-ritonavir.

Additionally, lopinavir C0 on study day 7 versus those on study day 10 and lopinavir C0 on study day 22 versus those on study day 24 were tested by the paired-samples t test to evaluate whether steady state was achieved. A power calculation was performed in the development phase of the study. The calculation, based on the lopinavir C0, indicated that data for nine subjects were needed in each study arm. As a dropout rate of 40 to 50% was assumed, 16 subjects were included in each study arm.

RESULTS

Subjects. Thirty-two subjects (18 males, 14 females) were included in the study, of which 20 completed the study. Twelve subjects dropped out for reasons of adverse events or laboratory abnormalities. Data for all 32 subjects participating in the study were included in the safety analyses. Pharmacokinetic data for 1 of the 20 subjects who completed the study were not evaluable due to vomiting shortly after drug administration on day 24. For this reason, statistical analyses for pharmacokinetics were performed with data for 19 subjects, 10 in arm 1 and 9 in arm 2.

The 10 subjects in arm 1 (4 males, 6 females) had a mean age of 37 years (range, 22 to 70 years), a mean height of 1.70 m (range, 1.61 to 1.85 m), and a mean weight of 70.6 kg (range, 61.5 to 77.0 kg). Of these 10 subjects, 1 was black; all others were Caucasian.

The nine subjects in arm 2 (7 males, 2 females) had a mean age of 36 years (range, 25 to 47 years), a mean height of 1.80 m (range, 1.58 to 1.90 m), and a mean weight of 75.4 (range, 60.5 to 85.4 kg). All nine subjects in arm 2 were Caucasian.

Lopinavir pharmacokinetics. Figure 1 shows the arithmetic mean ± standard deviation trough lopinavir levels in plasma in the morning obtained during the study. Trough lopinavir levels in plasma were not statistically different between days 7 and 10, suggesting that in both arms steady state was reached after 10 days of treatment with lopinavir-ritonavir at 400/100 mg BID (P = 0.5 and P = 0.5 for comparison of C0s on day 7 versus that on day 10 for arms 1 and 2, respectively). On study day 16 (when lopinavir-ritonavir at 400/100 mg BID was combined with rifampin at 600 mg QD), trough lopinavir levels decreased 93% in arm 1 and 90% in arm 2 in comparison to those on study day 10. Trough lopinavir levels increased in both study arms after lopinavir-ritonavir dosages were titrated to 800/200 mg BID in arm 1 and 400/400 mg BID in arm 2 and administered in combination with rifampin at 600 mg QD (days 18, 20, 22, and 24). Trough lopinavir levels in plasma were not statistically different between days 22 and 24, suggesting that steady state was reached (P = 0.7 and P = 0.15 for comparison of C0s
Although the geometric mean ratios are within 11% of unity for C_{\text{min}} and C_{\text{AUC}} on day 22 versus those on day 24 for both study days, the geometric mean ratio for C_{\text{AUC}} on day 22 is 91% compared to 81% on day 24. This difference is statistically significant. The arithmetic mean ratio for C_{\text{min}} on day 22 is 88%, whereas it was 81% on day 24. Figure 2 displays the arithmetic mean ratios for C_{\text{min}} and C_{\text{AUC}} for both study days 10 and 24. The error bars indicate standard deviations.

The values for the lopinavir pharmacokinetic parameters are summarized in Table 2. Arithmetic means ± standard deviations and geometric means are included for study days 10 and 24 for both study arms. The geometric means and error bars indicate standard deviations.

Although the geometric mean ratios are within 11% of unity for C_{\text{min}} and C_{\text{AUC}} on day 22 versus those on day 24 for both study arms, the geometric mean ratio for C_{\text{AUC}} on day 22 is 91% compared to 81% on day 24. This difference is statistically significant. The arithmetic mean ratio for C_{\text{min}} on day 22 is 88%, whereas it was 81% on day 24. Figure 2 displays the arithmetic mean ratios for C_{\text{min}} and C_{\text{AUC}} for both study days 10 and 24. The error bars indicate standard deviations.

The values for the lopinavir pharmacokinetic parameters are summarized in Table 2. Arithmetic means ± standard deviations and geometric means are included for study days 10 and 24 for both study arms. The geometric means and error bars indicate standard deviations.

Although the geometric mean ratios are within 11% of unity for C_{\text{min}} and C_{\text{AUC}} on day 22 versus those on day 24 for both study arms, the geometric mean ratio for C_{\text{AUC}} on day 22 is 91% compared to 81% on day 24. This difference is statistically significant. The arithmetic mean ratio for C_{\text{min}} on day 22 is 88%, whereas it was 81% on day 24. Figure 2 displays the arithmetic mean ratios for C_{\text{min}} and C_{\text{AUC}} for both study days 10 and 24. The error bars indicate standard deviations.
between the values of the pharmacokinetic parameters obtained on days 10 and 24 were observed. The total variability (coefficients of variation) in lopinavir \(C_{\text{min}}\) in arm 2 on days 10 and 24 were 36 and 46%, respectively. Figure 2 displays the arithmetic mean plasma-concentration time profiles for lopinavir in arm 2 for both study day 10 and study day 24. The error bars in Fig. 2 represent the standard deviation of the mean and show variabilities of the same magnitude on both study days.

**Ritonavir pharmacokinetics.** The values of the steady-state pharmacokinetic parameters for ritonavir are summarized in Table 3. Arithmetic means ± standard deviations and geometric means are included for study days 10 and 24 for both arm 1 and arm 2. The \(P\) values for the within-subject differences between days 10 and 24 are displayed as well. The geometric mean ratio (day 24/day 10) and 90% confidence interval are between days 10 and 24 are displayed as well. The geometric mean pharmacokinetic parameters for ritonavir on day 24 are presented in Table 3. Arithmetic means ± standard deviations (geometric means) unless indicated otherwise.

<table>
<thead>
<tr>
<th>Study arm and day</th>
<th>Arm 1</th>
<th>Arm 2</th>
<th>Day 10</th>
<th>Day 24</th>
</tr>
</thead>
<tbody>
<tr>
<td>AUC 12 (mg liter/h)</td>
<td>45.5 ± 19.2 (10.5)</td>
<td>120 ± 46 (10.5)</td>
<td>41.5 ± 16.1 (10.5)</td>
<td>142 ± 7 (10.5)</td>
</tr>
<tr>
<td>(C_{\text{max}}) (mg liter)</td>
<td>6.6 ± 2.9 (2.5)</td>
<td>18.3 ± 6.6 (2.5)</td>
<td>6.6 ± 2.6 (2.5)</td>
<td>18.3 ± 6.6 (2.5)</td>
</tr>
<tr>
<td>(t_{1/2}) (h)</td>
<td>0.67 ± 0.09 (0.27)</td>
<td>0.73 ± 0.09 (0.27)</td>
<td>0.67 ± 0.09 (0.27)</td>
<td>0.73 ± 0.09 (0.27)</td>
</tr>
<tr>
<td>(C_{\text{min}}) (mg liter)</td>
<td>0.05 ± 0.04 (0.14)</td>
<td>0.16 ± 0.04 (0.14)</td>
<td>0.05 ± 0.04 (0.14)</td>
<td>0.16 ± 0.04 (0.14)</td>
</tr>
<tr>
<td>(C_{\text{ave}}) (mg liter)</td>
<td>0.98 ± 0.57 (0.15)</td>
<td>1.28 ± 0.68 (0.15)</td>
<td>0.98 ± 0.57 (0.15)</td>
<td>1.28 ± 0.68 (0.15)</td>
</tr>
</tbody>
</table>

**Rifampin pharmacokinetics.** The values of the steady-state pharmacokinetic parameters for rifampin on day 24 are pre-
sent in Table 4. Arithmetic means ± standard deviations and geometric means are displayed. Data are grouped by study arm; arm 1 denotes lopinavir-ritonavir at 800/200 mg BID in combination with rifampin at 600 mg QD, and arm 2 denotes lopinavir-ritonavir at 400/400 mg BID in combination with rifampin at 600 mg QD. No statistically significant differences in rifampin pharmacokinetics between arms 1 and 2 were found.

**Adverse events.** Most (87%) of the adverse events were mild. Three adverse events (not related to a study medication) were reported to be severe; these were cases of gastroenteritis, influenza, and headache. Serious adverse events did not occur.

Twelve of 32 subjects (38%) were prematurely discontinued from the study; 3 of these subjects discontinued the study prior to randomization (while receiving lopinavir-ritonavir at 400/100 mg BID alone). One subject was prematurely discontinued from the study for grade 2 total bilirubin level elevations (>31 μmol/liter), which predominantly consisted of indirect bilirubin. However, this subject did not have concurrent grade 2 ALT, AST, or alkaline phosphatase level elevations. One additional subject was prematurely discontinued from the study for grade 3 elevations in cholesterol levels (>7.77 mmol/liter) and triglyceride levels (>8.48 mmol/liter), both of which subsequently declined to below grade 3 elevations following discontinuation of the study medication. A third subject was prematurely discontinued from the study for a complex of vomiting and abdominal pain. These complaints disappeared after the study medication was discontinued. There were half as many subject discontinuations in arm 1 (lopinavir-ritonavir at 800/200 mg BID and rifampin at 600 mg QD; three subjects discontinued the medication; one subject developed grade 2 elevations in AST, ALT, and GGT levels; one subject developed a grade 2 elevation in AST levels and a grade 3 elevation in ALT levels; and one subject suffered from vomiting and diarrhea) as in arm 2 (lopinavir-ritonavir at 400/400 mg BID with rifampin at 600 mg QD; six subjects discontinued the medication; three subjects developed grade 2 elevations in AST and ALT levels; two subjects developed grade 2 elevations in ALT levels; and one subject suffered from nausea, abdominal pain, fatigue, shivers, and increased sweating).

During the study, six subjects (two in arm 1, four in arm 2) had grade 2 elevations in ALT levels (>2.6 times the ULN) and three subjects (one in arm 1, two in arm 2) had grade 3 elevations in ALT levels (>5.1 times the ULN). Five of these subjects experienced concurrent grade 2 elevations in AST levels (>2.6 times the ULN). Seven of the nine subjects with grade 2 to 3 elevations in ALT levels (two in arm 1, five in arm 2) were prematurely discontinued from the study. The other two subjects developed elevations in liver enzyme levels on or after study day 24.

The onset of all grade 2 or 3 elevations in ALT and AST levels was after the initiation of rifampin treatment, but none of these were associated with grade 2+ elevations in total bilirubin or alkaline phosphatase levels. After discontinuation of the study medication, all such elevations declined below those for grade 2 toxicity, with only two remaining above the ULN at the final study evaluation.

**Laboratory measurements.** The mean change from the baseline values to the maximum values as well as the mean change from the baseline values to the final values was determined for a number of laboratory parameters. The baseline was day 1 for the period from day 1 to 10, and the baseline was day 10 for the period from days 11 to 24. For AST, the mean changes from the baseline value (day 10) to the maximum value (final value) were 39.9 (2.4) and 39.9 (1.1) IU/liter in arms 1 and 2, respectively, for study days 11 to 24. For ALT, the mean changes from the baseline value (day 10) to the maximum value (final value) were 72.6 (13.5) and 89.3 (6.9) IU/liter in arms 1 and 2, respectively, for study days 11 to 24. For alkaline phosphatase, the mean changes from the baseline value (day 10) to the maximum value (final value) were 12.9 (3.0) and 16.3 (5.0) IU/liter in arms 1 and 2, respectively, for study days 11 to 24. No clinically relevant changes in ASAT, ALT, and alkaline phosphatase levels were seen on study days 1 to 10 (lopinavir-ritonavir at 400/100 mg BID alone). For total bilirubin, the mean change from the baseline value (day 1) to the maximum value (final value) was 10 (4.7) μmol/liter for study days 1 to 10. For study days 11 to 24, the mean changes in the total bilirubin level from the baseline value (day 10) to the maximum value (final value) were 0.9 (−9.1) and 1.3 (−3.9) μmol/liter in study arms 1 and 2, respectively.

**DISCUSSION**

In the present study, two adjusted-dose regimens of lopinavir-ritonavir in combination with rifampin were compared to the standard dose of lopinavir-ritonavir without rifampin. The steady-state pharmacokinetics of lopinavir and ritonavir were determined after 10 days of treatment with the standard dose of lopinavir-ritonavir (400/100 mg BID). In the second part of the study, the steady-state pharmacokinetics of lopinavir-ritonavir at 800/200 mg BID with rifampin at 600 mg QD (arm 1) and lopinavir-ritonavir at 400/400 mg BID with rifampin at 600 mg OD (arm 2) were assessed.

**Lopinavir.** Lopinavir exposure was substantially higher in both study arms compared to the historical data obtained for lopinavir-ritonavir at 400/100 mg BID in combination with rifampin at 600 mg OD. This historical interaction study with a standard dose of lopinavir-ritonavir with rifampin was conducted with 22 healthy subjects to assess the effects of multiple

**TABLE 4. Steady-state pharmacokinetics of rifampin on study day 24**

<table>
<thead>
<tr>
<th>Arm</th>
<th>AUC12 (mg · h/liter)</th>
<th>Cmax (mg/liter)</th>
<th>Tmax (h)</th>
<th>t1/2 (h)</th>
<th>CL/F · kg (liter/h · kg)</th>
<th>V/F · kg (liter/kg)</th>
</tr>
</thead>
<tbody>
<tr>
<td>Arm 1</td>
<td>79.2 ± 33.84 (72.2)</td>
<td>14.2 ± 5.61 (13.0)</td>
<td>4.0 (3.0–4.0)</td>
<td>3.1 ± 1.75 (2.8)</td>
<td>0.12 ± 0.06 (0.11)</td>
<td>0.45 ± 0.16 (0.43)</td>
</tr>
<tr>
<td>Arm 2</td>
<td>76.6 ± 31.87 (70.3)</td>
<td>15.0 ± 3.80 (14.5)</td>
<td>4.0 (3.0–4.0)</td>
<td>2.5 ± 1.33 (2.2)</td>
<td>0.11 ± 0.06 (0.10)</td>
<td>0.34 ± 0.10 (0.33)</td>
</tr>
</tbody>
</table>

* Arm 1 (n = 10), lopinavir-ritonavir at 800/200 mg BID; arm 2 (n = 9), lopinavir-ritonavir at 400/400 mg BID.
* Values are arithmetic means ± standard deviations (geometric means) unless indicated otherwise.
* Values are medians (interquartile ranges).

On November 2, 2012 by Universiteitsbibliotheek

http://aac.asm.org/
doses of rifampin at 600 mg QD on the pharmacokinetics of lopinavir after treatment with lopinavir-ritonavir at 400/100 mg BID. The values of the pharmacokinetic parameters for lopinavir were substantially reduced by the coadministration of lopinavir-ritonavir with rifampin, as follows: $C_{\text{max}}$ by 45%, AUC by 75%, and $C_{\text{min}}$ by 99% (Bertz et al., 5th Int. Congr. Drug Ther. HIV Infect., abstr. P291, 2000).

While the concentrations of lopinavir are dramatically increased when lopinavir-ritonavir is used in combination with rifampin with both of the dosing regimens, it could not be demonstrated that the adjusted-dose regimens with rifampin evaluated in the present study were equivalent, particularly with respect to $C_{\text{min}}$, to the standard dose of lopinavir-ritonavir without rifampin. This indicates that the adjusted-dose regimens may in some cases not be capable of completely compensating for the accelerated metabolism of lopinavir by rifampin. Data are also limited by the relatively small number of subjects who completed the study and for whom pharmacokinetic data were evaluable. However, the level of lopinavir exposure in arm 2 was more comparable to that obtained with a standard dose lopinavir-ritonavir without rifampin than was the level of lopinavir exposure in arm 1. The pharmacokinetics of lopinavir in arm 1 were more variable than those in arm 2. It could be that the higher dose of ritonavir given in arm 2 (400 mg BID) resulted in a more consistent inhibition of lopinavir metabolism compared to that achieved with the ritonavir dose given in arm 1 (200 mg BID). Note that on study day 24, a total of 4 of 10 subjects (40%) in arm 1 had a $C_{\text{min}}$ lower than the lowest value observed on study day 10 ($C_{\text{min}} < 3.7$ mg/liter). In contrast, only one of the nine subjects (11%) in arm 2 had a $C_{\text{min}}$ lower than the lowest value observed on study day 10 ($C_{\text{min}} < 3.1$ mg/liter). The study was not designed to show a difference in lopinavir exposures between study arms. However, by taking into account the greater variability in lopinavir pharmacokinetics in arm 1, therapeutic drug monitoring might prove to be useful in clinical practice to monitor for possible subtherapeutic $C_{\text{min}}$s of lopinavir in plasma and individually optimize the lopinavir-ritonavir dosing regimen in a given patient. In arm 2, the $C_{\text{min}}$s of lopinavir in plasma tended to be higher, possibly making therapeutic drug monitoring of less importance.

**Ritonavir.** For ritonavir, it is apparent that in arm 1 a two-fold increase in the ritonavir dose from 200 mg/day in the absence of rifampin to 400 mg/day in the presence of rifampin resulted in a lesser than proportional increase in plasma ritonavir concentrations. In arm 2, considerably higher plasma ritonavir concentrations were achieved during treatment with lopinavir-ritonavir at 400/400 mg BID in combination with rifampin compared to those achieved with lopinavir-ritonavir at 400/100 mg BID alone. In fact, when ritonavir is administered in combination with lopinavir, the ritonavir $C_{\text{max}}$ and AUC_{12} increase more than proportionally due to nonlinear pharmacokinetics (8) when the total daily dose is increased fourfold from 200 mg/day in the absence of rifampin to 800 mg/day in the presence of rifampin. The ritonavir AUC_{12} was approximately fourfold higher when lopinavir-ritonavir at 400/400 mg BID was coadministered with rifampin than when lopinavir-ritonavir at 800/200 mg BID was coadministered with rifampin. These observed effects of ritonavir exposure indicate that the inhibition of CYP3A by ritonavir is more complete and less subject to induction by rifampin when ritonavir is dosed at 400 mg BID than when it is dosed at 200 mg BID.

**Rifampin.** Data for rifampin in the literature (2) report a mean $C_{\text{max}}$ and a mean AUC of 8 to 20 mg/liter and 60 to 80 mg · h/liter, respectively. The mean values for $C_{\text{max}}$ and AUC observed in this study are within these ranges (Table 4). This indicates that lopinavir-ritonavir does not affect the pharmacokinetics of rifampin. The fact that no statistically significant differences in the pharmacokinetic parameters for rifampin were observed between arm 1 and arm 2, as shown in Table 4, provides further evidence that these different doses of lopinavir and ritonavir had no influence on rifampin exposure. The literature also reports (2, 12) that rifampin intake with food can decrease the rifampin $C_{\text{max}}$. It is noteworthy that in the present study rifampin was administered with lopinavir-ritonavir at breakfast (550 kcal, 28% fat). Nevertheless, in this study the rifampin $C_{\text{max}}$ did not show the decrease that has been reported before. Data in the literature indicate that the $T_{\text{max}}$ is 1.5 to 2.0 h under fasting conditions. In a trial studying the single-dose pharmacokinetics of rifampin under fasting conditions (12), with food, and with antacids, the observed $T_{\text{max}}$ was 4.43 h after a high-fat breakfast (792 kcal, 57% fat). In the present study, the median $T_{\text{max}}$ was about 4 h in both study arms; this delay of $T_{\text{max}}$ was probably the result of the intake with food. However, in clinical practice, the rifampin $C_{\text{max}}$ is the main pharmacokinetic parameter of interest (2). Therefore, the clinical relevance of the delay in $T_{\text{max}}$ is limited, as in the present study the mean $C_{\text{max}}$s were well within the previously reported ranges (2).

**Safety.** The most common adverse events, reported by >50% of subjects, included urine discoloration, which is a known effect of rifampin therapy (5); nausea; headache; diarrhea; abdominal pain and cramps; and fatigue. The majority of all adverse events were mild (87%), with approximately 13% judged to be of moderate severity and only three events (gastroenteritis, influenza, and headache) reported to be severe. None of the adverse events met the regulatory definition of serious. A number of subjects had to discontinue the study prematurely due to elevations in liver function test results, with the onset of the elevations occurring after the initiation of combination lopinavir-ritonavir and rifampin dosing. A greater number of discontinuations occurred among the subjects in the arm receiving lopinavir-ritonavir at 400/400 mg BID plus rifampin at 600 mg QD. However, the study design did not allow an assessment of whether the frequency or magnitude of the elevations in the liver function test results seen with lopinavir-ritonavir in combination with rifampin was different between the study arms. No clinically significant hematology or urinalysis values were observed in the study.

Overall, tolerability limitations were observed with the coadministration of lopinavir-ritonavir and rifampin in healthy subjects. The high rate of discontinuations observed was primarily a result of the elevations in the liver function test results that occurred after the initiation of lopinavir-ritonavir and rifampin coadministration. However, there was no dosing segment with rifampin alone to allow determination of whether the liver function test abnormalities observed during combination lopinavir-ritonavir and rifampin treatment were of a greater magnitude or incidence than would have been observed with rifampin administration alone to healthy subjects.
Nevertheless, the increased rate of elevations in hepatic transaminase levels seen in both combination-treatment arms warrants the use of caution when these two drugs are administered concurrently to patients infected with both HIV and *M. tuberculosis*.

**Conclusions.** The present recommendations from the Centers for Disease Control and Prevention indicate that rifampin can be used in conjunction with efavirenz at 800 mg QD (4). The combination of efavirenz with rifampin has been studied in a group of 24 HIV-infected patients coinfected with *M. tuberculosis* (10). Other combination regimens that have been considered for use for the simultaneous treatment of HIV and *M. tuberculosis* infections are limited.

The combination of saquinavir-ritonavir at 400/400 mg BID with rifampin, as mentioned in the Introduction, has its parallels with the combination evaluated in the present study. However, the data for the saquinavir-ritonavir combination were only presented as a case report.

The product Trizivir combines three NRTIs, namely, zidovudine, lamivudine, and abacavir, and could be an option for use in combination with rifampin. Nevertheless, the pharmacokinetics of this combination of NRTIs in combination with rifampin were not studied, and recently, this combination of NRTIs was shown to be less effective than an efavirenz-based regimen (7) and therefore will not be an option of first choice.

Centers for Disease Control and Prevention guidelines also suggest the use of rifabutin (3, 4). However, complex bidirectional interactions are to be expected when rifabutin is combined with PIs (2). To compensate for these interactions, rifabutin doses must be decreased in some cases, or the PIs should be given at higher doses to compensate for accelerated metabolism. The combination of rifabutin with the NNRTIs efavirenz and delavirdine results in pharmacokinetic interactions as well (4); however, nevirapine can be used in combination with rifabutin, although no data from clinical studies have been published (4). It was reported from a study with HIV-infected inmates during a tuberculosis outbreak in a prison that, regardless of the rifabutin dosage, rifabutin concentrations are highly unpredictable, probably due to drug-drug interactions (16). From these data it becomes clear that the combination of rifabutin with PIs or NNRTIs remains a therapeutic challenge.

The presently studied combination of lopinavir-ritonavir, dosed as either 800/200 mg BID or 400/400 mg BID with rifampin, may be considered for the treatment of HIV-infected persons who are coinfected with *M. tuberculosis*. The toxicity observed when higher-dose lopinavir-ritonavir and rifampin were administered together led to discontinuation in 31% (9 of 29) of the healthy subjects during this study. When these drugs are used to treat patients who use other hepatically metabolized drugs, who are receiving long-term chronic treatment, and who have concomitant disease, the adverse event profile observed in the present study might even worsen. Therefore, the treatment of HIV-infected patients with tuberculosis with these agents should be approached with caution, and close monitoring of liver function will be needed. Therapeutic monitoring of the pharmacokinetics of lopinavir may be useful for the detection of minimal levels in plasma that are markedly below the expected mean, particularly in those patients treated with 800/200 mg BID, as well as to optimize the dosing regimen of lopinavir-ritonavir in combination with rifampin. If therapeutic drug monitoring is not possible, the combination of lopinavir and ritonavir at 400/400 mg BID may be preferred, although the rates of elevations in liver function test results achieved with that regimen tended to be higher than those achieved with the lopinavir-ritonavir regimen of 800/200 mg BID.

**ACKNOWLEDGMENTS**

We thank the subjects who participated in this study and Alina Bergshoeff, Jackie Droste, Marga de Graaff, Rosan Rosmalen, and the laboratory technicians of the Department of Clinical Pharmacy for help during performance and analysis of the results of this study. Kevin Niemi of the Antiviral Global Team at Abbott Laboratories, Abbott Park, IL, is thanked for logistical and operational support to the study report.

This study was funded by a grant from Abbott Laboratories.

**REFERENCES**


