Effect of Tipranavir-Ritonavir on Pharmacokinetics of Raltegravir

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Raltegravir (RAL; Isentress [also known as MK-0518]; Merck & Co., Inc., Whitehouse Station, NJ) is a novel human immunodeficiency virus (HIV) type 1 (HIV-1) antiretroviral agent that inhibits the enzyme responsible for catalyzing the stepwise process resulting in the integration of the HIV-1 DNA into the genome of the host cell (2, 4, 6; Isentress [RAL] package insert, Merck & Co., Inc.). RAL has potent in vitro activity against HIV-1, exhibiting a mean 95% inhibitory concentration of 31 nM in the presence of normal human serum (Isentress [RAL] package insert, Merck & Co., Inc.). In trials with viremic patients, RAL at dosages of 200 to 600 mg twice daily showed efficacy in reducing the HIV load below the threshold of reliable quantification in both treatment-naive patients (11) and heavily treated, multidrug-resistant patients (5). RAL at 400 mg is marketed in several countries for use in combination with other antiretroviral agents for the treatment of HIV-1 infection in treatment-experienced adult patients with evidence of viral replication and infection with HIV-1 strains resistant to multiple antiretroviral agents.

Dosing recommendations for RAL include coadministration with other active anti-HIV drugs, including tipranavir (TPV; Aplitiva; Boehringer Ingelheim International, Ridgefield, CT), an HIV-1 protease inhibitor (Aplitiva [TPV] package insert, Boehringer Ingelheim Pharmaceuticals, Inc.). TPV is both a substrate and an inducer of cytochrome 3A (CYP3A) (10; Aplitiva [TPV] package insert, Boehringer Ingelheim Pharmaceuticals, Inc.) and is a CYP1A2, CYP2C9, CYP2C19, and CYP2D6 inhibitor; a P-glycoprotein substrate; a weak P-glycoprotein inhibitor; and a potent P-glycoprotein inducer. TPV is recommended to be coadministered with ritonavir (RTV; Norvir; Abbott Laboratories, North Chicago, IL), a CYP3A inhibitor used to boost plasma TPV concentrations.; the net effect of TPV-RTV is both CYP3A inhibition and P-glycoprotein induction (10; Aplitiva [tipranavir] package insert, Boehringer Ingelheim Pharmaceuticals, Inc.; Norvir [RTV] package insert, Abbott Laboratories).

The metabolism of RAL is mediated by UDP-glucuronosyltransferase (UGT), primarily UGT1A1 (9). It is unknown whether TPV inhibits or induces glucuronosyltransferase; however, CYP3A, P-glycoprotein, and UGT1A1 are all regulated through the pregnane X receptor. Therefore, it is possible that TPV-RTV might also be an inducer of UGT1A1 and that the coadministration of TPV-RTV with RAL might affect the pharmacokinetics of RAL. RAL itself does not appear to appreciably induce or inhibit the enzymes involved in drug metabolism (9) and would therefore not be expected to affect the pharmacokinetics of TPV or RTV. The effect of the coadministration of multiple doses of RAL with RTV alone on the pharmacokinetics of RAL was examined in a previous study. Those analyses suggested that concomitant treatment with RTV had no clinically meaningful effect on the pharmacokinetics of RAL (8). This study was performed to assess the effect of the coadministration of multiple doses of TPV-RTV with multiple doses of RAL on the pharmacokinetics of RAL and to evaluate the safety and tolerability of the combination. (Portions of data presented here were printed in an abstract for the 46th Interscience Conference on Antimicrobial Agents and Chemotherapy [14].)
MATERIALS AND METHODS

Subjects. Healthy men and women between the ages of 18 and 50 years and within 30% of their ideal body weight were eligible for participation.Subjects were excluded if they were smokers, had any clinically significant medical history, had experienced recent excessive blood loss, or anticipated needing any prescription or nonprescription drugs during the conduct of the study. Women of child-bearing potential were required to have a negative beta-human chorionic gonadotropin pregnancy test.

Written informed consent was obtained from all subjects prior to study entry. The protocol was approved by the CRCC Independent Review Board in Austin, TX, and was conducted according to principles based on those of the Declaration of Helsinki.

Study design. This study was an open-label, three-period, fixed-sequence trial. During period 1, all subjects were administered oral RAL at 400 mg twice daily for 4 days, except that on day 4 only the morning dose was administered. During period 2, the same subjects were administered TPV at 500 mg plus RTV at 200 mg orally twice daily for 7 days. During period 3, all subjects received a combination of TPV-RTV and RAL at the doses administered during periods 1 and 2 twice daily for 4 days, except that no evening dose was given on day 4. Because the duration of period 2 was sufficient for the complete washout of RAL and any residual RAL would not be expected to affect the steady-state plasma concentrations of TPV or RTV, a washout interval between periods 1 and 2 was not required. There was no washout period between periods 2 and 3. The study drug was administered after a standardized moderate-fat meal, as recommended for TPV administration, except on pharmacokinetic sampling days, when the subjects received the study drug in the fasted state in order to reduce pharmacokinetic variability.

Although food increases the bioavailability and, hence, the level of exposure to TPV-RTV, the effect of fasting on sampling days is thought to be negligible because the induction of relevant enzymes would have already been established.

The safety and tolerability of RAL and TPV-RTV were assessed by clinical evaluation of vital signs, physical examinations, electrocardiograms, and laboratory safety evaluations (including hematology, serum chemistry, and urinalysis). The subjects were monitored for adverse experiences throughout the study.

Pharmacokinetic sampling and assays. Whole blood was collected for plasma RAL assay predosing on days 1 through 3 of period 1 and predosing and at 0.5, 1, 1.5, 2, 3, 4, 5, 6, 8, 10, and 12 h postdosing on day 4 of period 1. Samples were also collected predosing on days 2 and 3 of period 3 and predosing and at 0.5, 1, 1.5, 2, 3, 4, 5, 6, 8, 10, and 12 h postdosing on day 4 of period 3. Plasma samples were analyzed by a validated reverse-phase high-pressure liquid chromatography tandem mass spectrometry method (12). The lower limit of quantitation was 2 ng/ml (4.5 nM), and the linear calibration range was 2 to 1,000 ng/ml.

Pharmacokinetic methods. Plasma RAL concentrations, converted into molar units (nM) by using the molecular weight of 444.4, and actual sampling times, converted to the elapsed time relative to the RAL dosing times, were used to estimate the pharmacokinetic parameters (with the exception of the time to the maximum concentration of drug in plasma [Tmax]) for each treatment in each subject. Values below the limit of quantitation (BLQ) of the plasma assay, i.e., <2 ng/ml (<4.5 nM), were replaced according to the following rules: the predose BLQ value was 0; the first BLQ value in the terminal phase was (1/2) · the lower limit of quantitation, which was 1 ng/ml (2.3 mM); and second and subsequent BLQ values in the terminal phase were 0. The area under the concentration-time curve (AUC) from time zero to 12 h (AUC0–12) was calculated by using the linear trapezoidal method for ascending concentrations and the log trapezoidal method for descending concentrations. The maximum concentration of drug in plasma (Cmax) and Tmax were obtained by inspection. Nominal plasma sampling times were considered to be the next time point after the actual observed Tmax if it did not differ meaningfully from the nominal plasma sampling time. The concentrations at 12 h (C12) were assessed from the plasma concentrations determined for the nominal sampling times at 12 h postdosing.

Statistical analysis. The pharmacokinetic parameter values C12, AUC0–12, and Cmax were natural-log transformed before analysis. As such, all corresponding 90% confidence intervals (CIs) for means (difference of two means) were constructed on the natural-log scale. Exponentiation was performed on the means (mean differences) and lower and upper limits of these CIs. With the exception of Tmax, all CIs were based on an analysis of variance (ANOVA) model with treatment as a fixed effect and with subject as a random effect (a compound symmetric covariance structure was assumed). Only C12 arising from the final dosing interval (day 4) of each period were included in the ANOVA model. For Tmax, the Hodges-Lehman estimate of the true median difference was computed (RAL with TPV-RTV minus RAL alone), as was a 90% CI for this value.

RESULTS

Demographics and baseline characteristics. Eighteen subjects (7 women, 11 men; 2 African-American subjects) were enrolled in the study. The mean age was 30.2 years (range, 19 to 47 years), and the mean weight was 79.9 kg (range, 56.2 to 108.2 kg). The subjects were HIV negative and in good general health, according to a routine medical history, physical examination, vital signs, and laboratory data. All 18 subjects were included in the analysis of safety. Fifteen subjects completed the study per protocol and were included in the pharmacokinetic analyses. Of the three subjects who discontinued the study, two subjects withdrew consent and one was withdrawn due to non-drug-related adverse experiences.

RAL pharmacokinetics. The arithmetic mean RAL plasma concentration-time profiles for the administration of RAL alone and with TPV-RTV are shown in Fig. 1, and a summary of the RAL pharmacokinetic parameter values is provided in Table 1. The geometric mean ratio (RAL with TPV-RTV/RAL alone) for RAL C12 was 0.45 (90% CI, 0.31, 0.66) (P = 0.0021). The coadministration of RAL with TPV-RTV modestly decreased the RAL AUC0–12 (−24%) and Cmax (−18%) compared to the values obtained by the administration of RAL alone. The RAL Tmax was not substantially affected. Figure 2 displays the individual treatment ratios, the treatment geometric mean ratio (GMR), and the 90% CIs for RAL C12 (Fig. 2a) and Cmax (Fig. 2b).

Safety and tolerability. RAL was generally well tolerated. No serious clinical or laboratory adverse experiences were reported. Three of the 18 subjects enrolled in the study discontinued prior to the completion of the study. Two of these subjects withdrew consent. The third subject discontinued because of non-drug-related adverse experiences suffered in a motor vehicle accident.

A total of 33 clinical adverse experiences were reported by 17 subjects. Eighteen of the 33 clinical adverse experiences, reported by 13 of the subjects, were judged by the investigator to be related to the study drug. All drug-related adverse experiences were mild to moderate in intensity and transient in
nature. The most common drug-related adverse experiences (reported by more than one subject) were nausea ($n = 5$), headache ($n = 2$), and loose stools ($n = 2$). No laboratory adverse experiences were reported.

**DISCUSSION**

Drug-drug interactions are an important consideration in the treatment of HIV-infected populations because they often affect the choice of antiretroviral agent and other agents to be used as well as adherence to the treatment regimen. In this study, we examined the effect of coadministration of multiple doses of RAL together with multiple doses of TPV-RTV on the pharmacokinetics of RAL. Multiple doses of TPV-RTV decreased the RAL $C_{12}$; the $C_{12}$ treatment GMR (RAL with TPV-RTV/RAL alone) was 0.45, and the 90% CI for the GMR was 0.31 to 0.66. The ranges of RAL $C_{12}$ with and without TPV-RTV were 14 to 624 nM and 57 to 416 nM, respectively, with a corresponding GMR range of 0.17 to 4.05. These ranges suggest that the pharmacokinetic variability observed in this study was relatively high. In contrast to $C_{12}$, the effects on $AUC_{0-12}$, $C_{\text{max}}$, and $T_{\text{max}}$ were less pronounced. RAL is eliminated via glucuronidation, which is predominantly mediated by the enzyme UGT1A1. The effects on the RAL $C_{12}$ are consistent with the induction of glucuronidation; however, the lack of a substantive effect on $AUC_{0-12}$ or $C_{\text{max}}$ suggests that the overall inductive effect is minor.

The effect of the coadministration of multiple doses of RAL and RTV on the pharmacokinetics of RAL was examined in a previous study (8). Those analyses demonstrated that concomitant treatment with RTV had no clinically meaningful effect on the pharmacokinetics of RAL (8). Although the dosage of RTV differed between the previous and the current studies and the induction of UGT may be dose dependent, the decrease in $C_{12}$ is likely due to the inductive effects of TPV rather than the inductive effects of RTV. TPV has been shown to affect the activation of CYP enzymes and P-glycoprotein (Aptivus [TPV] package insert, Boehringer Ingelheim Pharmaceuticals, Inc.), and the data presented here support the likelihood that TPV induces glucuronidation as well. Because considerable in vitro and in vivo data suggest that RAL does not appreciably induce or inhibit the enzymes involved in drug metabolism (1, 7, 9) (including CYP3A4, the primary enzyme involved in the elimination of TPV [Aptivus {TPV} package insert, Boehringer Ingelheim Pharmaceuticals, Inc.] and RTV [Norvir {RTV} package insert, Abbott Laboratories]), the effects of RAL on the pharmacokinetics of TPV-RTV were not explored in this study.

The results of this study suggest that the RAL $C_{12}$ was decreased when RAL was coadministered with TPV-RTV. However, considerable clinical efficacy data are available from the RAL phase III program, in which approximately 100 patients received 400 mg RAL twice daily in combination

### Table 1. Comparison of plasma RAL pharmacokinetic parameter values in healthy subjects administered multiple doses of RAL at 400 mg twice daily alone and in combination with multiple doses of TPV-RTV twice daily

<table>
<thead>
<tr>
<th>Parameter</th>
<th>$C_{12}$ (nM)$^a$</th>
<th>$AUC_{0-12}$ (µM·h)$^b$</th>
<th>$C_{\text{max}}$ (µM)$^b$</th>
<th>$T_{\text{max}}$ (h)</th>
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</thead>
<tbody>
<tr>
<td>RAL alone</td>
<td>129 (84, 198)$^c$</td>
<td>9.97 (6.08, 16.35)</td>
<td>2.85 (1.48, 5.48)</td>
<td>4.0$^d$</td>
</tr>
<tr>
<td>RAL with TPV-RTV</td>
<td>59 (39, 89)</td>
<td>7.61 (4.64, 12.48)</td>
<td>2.33 (1.21, 4.48)</td>
<td>2.0$^d$</td>
</tr>
<tr>
<td>RAL with TPV-RTV/RAL alone</td>
<td>0.45 (0.31, 0.66)$^e$</td>
<td>0.76 (0.49, 1.19)</td>
<td>0.82 (0.46, 1.46)</td>
<td>−0.6 (−1.5, 0.3)$^e$</td>
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$^a$ The data are for 15 subjects, unless indicated otherwise.

$^b$ The data represent the geometric mean computed from least-squares estimate from an ANOVA performed on the natural-log-transformed values. The values in parentheses are the 95% CIs for RAL alone and RAL with TPV-RTV and the 90% CIs.

$^c$ n = 14 subjects.

$^d$ Median value.

$^e$ Hodges-Lehman estimate of median treatment difference and 90% CI for true median treatment difference.
with optimized background therapy (OBT), which included TPV-RTV (3, 13). At 24 weeks, the efficacy observed in this subgroup was comparable to that observed in subjects not receiving TPV-RTV. Specifically, the proportions of patients infected with TPV-susceptible virus (as determined by genotypic testing) who achieved HIV RNA loads of less than 50 copies/ml at 24 weeks and who used TPV in combination with OBT were 73% for RAL-treated patients and 40% for placebo-treated patients; the proportions were 36% and 15% for those infected with TPV-resistant virus who received TPV in combination with OBT and placebo-treated patients, respectively. For those patients who did not receive TPV in combination with OBT and placebo-treated patients, the proportions were 66% and 36%, respectively (3). In each subgroup, RAL in combination with OBT was associated with an observed treatment benefit of at least 20% over the benefit of placebo plus OBT. This observation supports the suggestion that TPV-RTV may be coadministered with RAL without a dose adjustment.

In conclusion, multiple doses of TPV-RTV decreased the RAL C_{12h}, although AUC_{0–12} and C_{max} were not substantially affected. Favorable efficacy data collected in phase III substudies that TPV-RTV may be coadministered with RAL without a dose adjustment. The coadministration of RAL and TPV-RTV was generally well tolerated.

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T. Champ was responsible for enrollment of the subjects, collection of the data, analysis and interpretation of the data, and preparation of the manuscript. A. Moreau and E. Mangin were responsible for analysis and interpretation of the data and preparation of the manuscript. K. M. Gottesdiener, W. D. Hanley, M. Iwamoto, J. T. Kost, J. A. Stone, J. A. Wagner, and L. A. Wenning were responsible for the study concept and design, analysis and interpretation of the data, and preparation of the manuscript.

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REFERENCES