Pharmacokinetic Interaction between Amprenavir and Clarithromycin in Healthy Male Volunteers

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Received 14 April 1999/Returned for modification 18 September 1999/Accepted 8 January 2000

The P450 enzyme, CYP3A4, extensively metabolizes both amprenavir and clarithromycin. To determine if an interaction exists when these two drugs are coadministered, the pharmacokinetics of amprenavir and clarithromycin were investigated in healthy adult male volunteers. This was a Phase I, open-label, randomized, balanced, multiple-dose, three-period crossover study. Fourteen subjects received the following three regimens: amprenavir, 1,200 mg twice daily over 4 days (seven doses); clarithromycin, 500 mg twice daily over 4 days (seven doses); and the combination of the above regimens over 4 days (seven doses of each drug). Twelve subjects completed all treatments and the follow-up period. The erythromycin breath test (ERMBT) was administered at baseline, 2 h after the final dose of each of the three regimens and at the first follow-up visit.

Coadministration of clarithromycin and amprenavir significantly increased the mean amprenavir AUCₜₚ, Cₘₐₓ,ss, and Cₘᵢₙ,ss by 18, 15, and 39%, respectively. Amprenavir had no significant effect on the AUCₜₚ of clarithromycin, but the median Tₘᵢₙ,ss for clarithromycin increased by 2.0 h, renal clearance increased by 34%, and the AUCₜₚ for 14-(R)-hydroxyclarithromycin decreased by 35% when it was given with amprenavir. Amprenavir and clarithromycin reduced the ERMBT result by 85 and 67%, respectively, and by 87% when the two drugs were coadministered. The baseline ERMBT value did not correlate with clearance of amprenavir or clarithromycin. A pharmacokinetic interaction occurs when amprenavir and clarithromycin are coadministered, but the effects are not likely to be clinically important, and coadministration does not require a dosage adjustment for either drug.
Dextrose Injection, USP). On days 4, 8, and 12, the injection was given 2 h after
usual 1, 2, and 3 months after completion of the treatment phase. The occurrence
low-up visits to monitor LFTs were scheduled weekly until they resolved. If no
abnormalities were noted, then subjects were discharged from the study center
according to the product information from Metabolic Solutions Inc. Each subject
4, 8, and 12), and at the follow-up evaluation. The ERMBT was performed
establish a baseline), on the fourth dosing day of each treatment period (i.e., days
A sample for the analysis of clarithromycin or 14-(R)-hydroxyclarithromycin was drawn into a 5-ml presealed green-
 stopped VACUTAINER tube (containing sodium heparin). Each sample was centrifuged at 2000× g
+4°C to separate the plasma. Urine was collected predosing to establish a baseline and thereafter over the
intervals: 0 to 4, 4 to 8, 8 to 12, and 12 to 24 h postdosing on each of days 4, 8, and 12. For the predosing sample, subjects voided their bladders 15 min prior to dosing. For all postdosing collection intervals, subjects were allowed to
void their bladders as needed during and at the end of the collection interval.
Individual plasma and urine samples were aliquoted into propylene storage tubes, labeled, and stored upright in a non-self-defrosting freezer (–20°C or lower) until shipment to Glaxo Wellcome, Inc., for analysis of amprena-
by International Bioanalytical (Glaxo Wellcome) or to BAS Analytics, West
Lake, Ind., for analysis of clarithromycin or 14-(R)-hydroxyclarithromycin.
Plasma analytical methods. Plasma concentrations of amprenavir were deter-
mined by a semi-automated solid-phase extraction method. A 0.5-ml portion of
plasma was combined with 0.5 ml of internal standard solution (VB 11599, 5.0
µg/ml). Solid-phase extraction was performed with a Waters MilliLab Worksta-
tion and C18 Sep-Pak cartridges. The samples were loaded onto Waters C18
SPE cartridges (3 mm × 3 cm) by rotary column extraction. The cartridge con-
centrated the samples and eluted the drug using 2.5 ml of methanol, followed by water. After the calibration standard, the control or
sample was loaded, and the cartridge was washed with water and methanol
(65:35, vol:vol). The compound was eluted from the cartridges with 2.5 ml of acetonitrile. The volume of the eluate was reduced by evaporation under nitro-
gen at 37°C. A redisolved sample in the mobile phase was then loaded on the Waters Symmetry C18 column (3.5 by 150 mm) maintained at 40°C and eluted with a mobile phase consisting of acetonitrile-water in a 45:55 (vol:vol) ratio at a flow rate of 1.0 ml/min. The samples were injected in the range of 25–500 ng/ml (analytical range: 10–250
µg/ml). The amprenavir calibration standard concentra-
tions were linear from 10 to 1,000 ng/ml; the amprenavir plasma control con-
centrations were 30, 400, and 800 ng/ml. The clinical samples were diluted into the range of the calibration curve with blank human plasma and reasayed if they exceeded the upper limit of quantitation (1,000 ng/ml).
Upon validation of the amprenavir assay technique, the interassay precision,
assessed from spiked validation control samples (n = 6) at four concentrations
over four analytical runs with human plasma and expressed as percent coefficient of variation (CV), ranged from 1.8 to 7.7%. The intraassay precision ranged from 1.8 to 11.3%. The percent recovery of amprenavir was determined in human plasma at concentrations of 75, 400, and 800 ng/ml (n = 6 at each concentration) by injecting analytical standards (with internal standard) directly onto the col-
umn and calculating the recovery. The precision of the internal standard was based on the concentration range of 80 to 88% across the concentration range of 75 to 800 ng/ml.
Concentrations of clarithromycin and 14-(R)-hydroxyclarithromycin in plasma were determined by liquid chromatography-tandem mass spectrometry (LC-MS-MS). Clarithromycin, 14-(R)-hydroxyclarithromycin, and 14–(S)-hydroxyclarithromycin were extracted from 1.0
ml of heparinized plasma by liquid-liquid extraction at an alkaline pH. Erythro-
mycin B served as an internal standard. After the addition of carbonate solution
and internal standard to the plasma, the macroldes were extracted into methyl-
t-butyl ether. The ether layer was transferred to a clean tube and reconstituted
with a pH 6 buffer-acetonitrile mixture. The reconstituted extract was washed
with hexane and injected into an LC-MS-MS system with atmospheric pressure
chemical ionization. For clarithromycin and 14-(R)-hydroxyclarithromycin calibration standard concentrations ranged from 15.6 to 8,000 ng/ml, and the quality control concentra-
tions were 40, 400, and 1,000 ng/ml in human plasma. For the clarithromycin calibration standards, the interday CV was ±6.8%; the intra-day CV ranged from 5.3 to 9.9%. For the 14-(R)-hydroxyclarithromycin calibration standards, the interday CV was ±6.8%; the intra-day CV ranged from 1.7 to 7.4%. Standard curve correlation coefficients for both compounds were ≥0.985.
ERMBT analytical procedures. All ERMBT samples were assayed at the VCU
School of Pharmacy Biopharmaceutical Analysis Laboratory. Liquid scintillation
counting was used to measure radioactivity. Ten ml of scintillation cocktail (Packard Instrument Co.) was added to decolorized samples in scintillation vials; samples were mixed well and left in the dark at room temperature for at least 16 h. The samples were counted on a Packard Model Tri-Carb 4500 liquid scintillation counter with 4.0 μCi in 0.5 ml of 100% ethanol, USP, diluted in 4.5 ml of 5%
Dextrose Injection, USP. On days 4, 8, and 12, the injection was given 2 h after
administration of the seventh dose of each treatment, immediately after injec-
tion of the 2-h postdosing pharmacokinetic blood sample(s). Twenty minutes
after the injection, the subject exhaled through a plastic straw into 4 ml of benzethonium hydroxide-solution (a CO2-trapping agent) in 20-ml glass
scintillation vials until the color changed from blue to clear, indicating that 2 ml CO2 had been trapped. The time required for CO2 to be trapped muni-
factured 1 min. Each sample was tightly capped and stored at 4°C until assayed.
If, following completion of all postdosing procedures for day 12, no clinical
abnormalities were noted, then subjects were discharged from the study center
with instructions to return 7 to 10 days later for the first follow-up visit. If there
were significant elevations in liver function tests (LFTs), then subsequent fol-
low-up visits to monitor LFTs were scheduled weekly until they resolved. If no
significant LFT elevations were noted, subsequent follow-up visits were sched-
uled on days 21 and 28 following completion of the treatment phase. The occurrence
of adverse effects was monitored throughout the treatment phase of the study
and again at follow-up visits.
Pharmacokinetic analyses. The observed peak plasma drug concentrations at steady state ($C_{\text{max,ss}}$) and the time for each drug to reach peak concentrations ($T_{\text{max,ss}}$) were obtained by inspection of the individual plasma concentration-time data. The minimum drug concentration at steady state ($C_{\text{min,ss}}$) was calculated as $(C_0 + C_F)/2$, where $C_0$ is the plasma concentration before the last dose and $C_F$ is the plasma concentration of the last sample of the steady-state dosing interval. The AUC at steady state (AUCss), from the time of the predosing sample to the last sample of the steady-state dosing interval was calculated for each volunteer using the linear trapezoidal rule. The apparent total clearance at steady-state (CL/F) was calculated as dose/AUCss. Similar formulae were used to determine 14-($R$)-hydroxyclarithromycin pharmacokinetic parameters. The ratio of the metabolite AUC to the parent drug AUC (AUC$_{14-OH-clar}$/AUC$_{clar}$) was also calculated based on the AUCss.

Urine pharmacokinetic parameters were determined for clarithromycin and 14-($R$)-hydroxyclarithromycin only. Renal clearance (CLr) was calculated as $Ae_{ss}/AUC_{ss}$, where $Ae_{ss}$ is the amount of drug excreted in the urine over the dosing interval. The percentages of clarithromycin and its metabolite eliminated in the urine were calculated based on clarithromycin weight equivalents. The molecular sizes of clarithromycin and 14-($R$)-hydroxyclarithromycin were 747.96 and 763.96 Da, respectively.

The pharmacokinetic profiles obtained when the two drugs were administered together were compared with the profiles obtained when the drugs were administered alone (i.e., amprenavir plus clarithromycin versus amprenavir alone; amprenavir plus clarithromycin versus clarithromycin alone).

Statistical analysis. The primary analysis of pharmacokinetic parameters (other than $T_{\text{max,ss}}$) was performed after log transformation. Analyses of variance (ANOVA) considering sequence, period, and treatment as fixed effects and subject within sequence as the random effect, were performed using the Mixed Linear Models procedure (SAS PROC MIXED, version 6.12; SAS Institute, Cary, N.C.). The geometric least-squares mean and 90% confidence intervals (90% CI) were calculated for each pharmacokinetic parameter, along with their descriptive summary statistics. Two one-sided t tests (90% CI) were performed to compare the pharmacokinetic parameters obtained when the combination treatments were administered with those for drug given alone. The $T_{\text{max,ss}}$ was analyzed on a pairwise basis using a Wilcoxon signed rank test ignoring periods. Estimations of the median difference between treatments and 90% CI were calculated. Pearson’s correlation coefficient was calculated for potential linear relationships between continuous variables.

Descriptive statistics of ERMBT results at baseline, 2 h after dosing (days 4, 8, and 12) and at the first follow-up visit were summarized by calculation of the mean reduction in ERMBT compared with the baseline, and the respective 95% CI.

RESULTS

Study subjects. A total of 14 HIV-seronegative, healthy males (12 Caucasian and 2 African-American) were enrolled in this study. Thirteen subjects received all three treatments, but only 12 subjects completed all phases of the study. One subject was withdrawn midway through his second treatment (amprenavir plus clarithromycin) after complaining of nausea and vomiting. The other subject withdrew during the third treatment (amprenavir plus clarithromycin) for personal reasons.

Adverse events. There were no serious adverse events reported during this study, and all three treatments were generally well tolerated. The 14 subjects reported a total of 188 adverse events. The most common adverse events for amprenavir were mild gastrointestinal events (50%) and oral numbness (43%). Clarithromycin was most commonly associated with a bad taste (31%). Combination treatment with amprenavir plus clarithromycin resulted in greater subject intolerance than treatment with either drug alone, with any gastrointestinal events (71%) and oral numbness (50%) accounting for the majority of adverse effects. There was no apparent effect of the study drugs on hematology, clinical chemistry, or urinalysis laboratory values, nor any apparent changes in vital signs, physical examination findings, or electrocardiogram data from screening to follow-up.

Pharmacokinetics. (i) Amprenavir. Concentrations of amprenavir immediately before the final dose ($C_0$) were not different from concentrations 12 h after the final dose, indicating that steady state had been achieved. Figure 1 illustrates the effect of clarithromycin on mean plasma amprenavir concentrations. There were statistically significant increases in the amprenavir AUCss (18%), $C_{\text{max,ss}}$ (15%), and $C_{\text{min,ss}}$ (39%), and a decrease in CL/F (15%), when amprenavir was administered with clarithromycin (Table 1). There was a nearly significant negative correlation between the baseline amprenavir AUC and the percent change in the amprenavir AUCss when amprenavir was given with clarithromycin ($r^2 = 0.30; P = 0.065$). There was a significant negative correlation between the AUCss for clarithromycin and the magnitude of percent change from baseline in the amprenavir AUCss ($r^2 = 0.44; P = 0.02$). There was no significant association between subject weight and the AUCss for amprenavir ($r^2 = 0.24; P = 0.10$). The medians of $T_{\text{max,ss}}$ were not different between treatments.
There were no significant period or sequence effects in any of the ANOVA comparisons.

(ii) Clarithromycin. Concentrations of clarithromycin immediately before the final dose \((C_0)\) were not different from concentrations 12 h after the final dose, indicating that steady state had been reached. Amprenavir had no significant effect on the geometric least-squares means for the clarithromycin AUC\(_{ss}\), \(C_{\text{min,ss}}\), and CL/F (Fig. 2; Table 2). The median \(T_{\text{max,ss}}\) following administration of the combined treatment was 2.0 h later than that after the administration of clarithromycin alone \((P < 0.05)\). There was a 34% increase in CL\(_R\) with the combined treatment over that with clarithromycin alone. There was no significant linear correlation between the baseline apparent oral clearances for clarithromycin and amprenavir \((r^2 = 0.22; P = 0.11)\). Weight was able to explain a significant amount of variability in the AUC\(_{ss}\) for clarithromycin \((r^2 = 0.34; P = 0.04)\); larger subjects had a lower AUC\(_{ss}\).

(iii) 14-\((R)\)-Hydroxyclarithromycin. Figure 3 illustrates the effect of amprenavir on mean plasma 14-\((R)\)-hydroxyclarithromycin concentrations. A summary of the results for 14-\((R)\)-hydroxyclarithromycin parameters is presented in Table 3. Amprenavir clearly reduced the formation of the main metabolite for clarithromycin, resulting in statistically significant decreases in the 14-\((R)\)-hydroxyclarithromycin AUC\(_{ss}\) (35%) and \(C_{\text{max,ss}}\) (32%). There was a 37% decrease in the AUC\(_{14-OH-clar}/\text{AUC}_{clar}\) ratio. The median \(T_{\text{max,ss}}\) following administration of the combined treatment was 2.0 h later than that following the administration of clarithromycin alone. The percentage of the dose excreted in the urine as 14-\((R)\)-hydroxyclarithromycin was 16% lower with the combined treatment than with clarithromycin alone.

The AUC\(_{ss}\) for amprenavir given alone did not predict the magnitude of the percent reduction in the baseline AUC\(_{ss}\) for 14-\((R)\)-hydroxyclarithromycin \((r = 0.17; P = 0.61)\).

![FIG. 2. Mean plasma clarithromycin concentrations (± standard deviations) versus time (n = 12 subjects) when clarithromycin was given alone (solid circles) or coadministered with amprenavir.](http://aac.asm.org/)
ERMBT. The mean reduction in the ERMBT result was 85% (95% CI, 78 to 92%) after the administration of amprenavir, 67% (95% CI, 59 to 74%) for clarithromycin, and 87% (95% CI, 79 to 94%) for both drugs administered concurrently (Fig. 4). These data are consistent with evidence that drug interactions between clarithromycin and CYP3A4 substrates are of a lower magnitude compared with the effects of HIV-1 protease inhibitors (5). There was no significant correlation between the baseline ERMBT result and the C/F for amprenavir ($r = 0.30; P = 0.35$) or clarithromycin ($r = 0.28; P = 0.38$). There was a nearly significant negative correlation between the percent reduction in the ERMBT result following clarithromycin treatment and the percent reduction in the clearance of amprenavir ($r^2 = 0.34; P = 0.06$). The mean ERMBT result at follow-up (2.08% ± 0.63% metabolized/h) was not significantly different from baseline (2.31% ± 0.68% metabolized/h; $P = 0.107$).

**DISCUSSION**

The pharmacokinetics of amprenavir and clarithromycin when given alone are in agreement with the findings of previous investigations (3, 19). Clarithromycin given in combination with amprenavir resulted in statistically significant changes in selected pharmacokinetic parameters for both drugs. Clarithromycin increased the amprenavir AUC, $C_{\text{max,ss}}$, and $C_{\text{min,ss}}$ by 18, 15, and 39%, respectively, with an associated 15% decrease in CL/F. While this interaction is statistically significant, it is unlikely to be clinically important. An 18% increase in the AUC is within the intersubject variability normally seen when amprenavir, 1,200 mg every 12 h, is used clinically (19). In addition, the 39% mean increase in $C_{\text{min,ss}}$ is not likely to be a safety concern, since the absolute effect is small (mean increase from 0.38 to 0.53 μg/ml) and there is no known adverse event related to increased amprenavir trough concentrations.

Administration of amprenavir with clarithromycin had no statistically significant effect on the pharmacokinetic parameters AUC, $C_{\text{max,ss}}$, and $C_{\text{min,ss}}$ for clarithromycin. However, the AUC, $C_{\text{max,ss}}$, and $C_{\text{min,ss}}$ of 14-(R)-hydroxyclarithromycin were decreased 35 and 32%, respectively by amprenavir; there was a 28% increase in CL of this metabolite; and the CL of clarithromycin increased by 34%. This reduced formation of 14-(R)-hydroxyclarithromycin appeared to be balanced by increased CL of the parent drug, resulting in no net change in the AUC for clarithromycin. The metabolism of clarithromycin to 14-(R)-hydroxylaritromycin is mediated by CYP3A4 (18), and the decreases in the 14-(R)-hydroxyclarithromycin AUC and $C_{\text{max}}$ are consistent with inhibition of CYP3A4 by amprenavir. Ritonavir has a similar effect on the metabolism of clarithromycin, but of greater magnitude (15). The mechanism for increased CL of clarithromycin is unclear but is unlikely to represent protein-binding displacement, since clarithromycin is approximately 70% bound to albumin, and binding would have to decrease to nearly zero to account for the increase in CL. Furthermore, amprenavir has no known effects on renal function and should not alter renal secretion of clarithromycin. It is possible that the reduced formation of the metabolite may decrease competition with the parent compound for secretion, resulting in an increase in the CL of clarithromycin, but this remains conjectural.

It is unlikely that the changes in clarithromycin and 14-(R)-hydroxyclarithromycin pharmacokinetics are clinically relevant. 14-(R)-Hydroxylaritromycin has in vitro activity against...
Increased the AUC ss. Likewise, there was a significant negative correlation between the AUC ss of clarithromycin and the magnitude of per-

tance. Third, a number of correlation analyses are not con-

some bacterial pathogens and may contribute to the clinical efficacy of clarithromycin, especially for infections caused by Haemophilus influenzae (8), but is less important for MAC (13). While it is possible that the therapeutic efficacy of clari-

thromycin may be compromised as a result of this interaction, the effect of amprenavir is less than that of other protease inhibitors (Table 4). There are no published reports of therapeutic failure when clarithromycin has been used to treat bacterial infections in HIV-infected patients receiving protease inhibitors, and dosage adjustments are not recommended for patients receiving other protease inhibitors and clarithromycin.

We have attempted to determine a mechanism for these effects. Since erythromycin (in the ERMBT), clarithromycin, and amprenavir are at least partially metabolized by hepatic CYP3A4, we hypothesized that there would be significant correlations of metabolic parameters between these three drugs. However, the mechanism(s) of the interactions described above appears to be more complex than simple alterations in hepatic CYP3A4 metabolism, as suggested by a number of observations. First, a good correlation between the ERMBT result and clearance of a CYP3A substrate has been suggested as evidence that the substrate is largely metabolized by hepatic CYP3A (7). In contrast, we found that the ERMBT results at baseline did not predict clearance of either amprenavir or clarithromycin, which suggests that nonhepatic mechanisms are more relevant (below). Second, although both amprenavir and clarithromycin significantly reduce hepatic CYP3A4 activity as measured by the ERMBT, amprenavir caused significantly greater suppression than clarithromycin (Fig. 4). However, clarithromycin had a more pronounced effect on serum amprenavir concentrations than amprenavir had on serum clarithromycin concentrations, an effect opposite to that which would be expected if hepatic metabolism were of central import-

Table 4. Comparison of protease inhibitor effects on clarithromycin pharmacokinetics

<table>
<thead>
<tr>
<th>Agent</th>
<th>% Clarithromycin increase (90% CI)</th>
<th>14-OH-CLAR b decrease (90% CI)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>AUC</td>
<td>C max</td>
</tr>
<tr>
<td>Saquinavir (1,200 mg</td>
<td>45 (17–81)</td>
<td>39 (10–76)</td>
</tr>
<tr>
<td>every 8 h)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Ritonavir (200 mg</td>
<td>77 (56–103)</td>
<td>31 (15–51)</td>
</tr>
<tr>
<td>every 8 h)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Indinavir (800 mg</td>
<td>53 ± 36'</td>
<td>NS b</td>
</tr>
<tr>
<td>every 8 h)</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Amprenavir (1,200 mg</td>
<td>No effect</td>
<td>-10</td>
</tr>
<tr>
<td>BID)</td>
<td></td>
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* Results are expressed as percent change from values with clarithromycin alone.

Additional mechanisms that may explain the effects observed include alterations in CYP3A4-mediated gastrointestinal metabolism and alterations in P-glycoprotein (P-gp)-mediated gastrointestinal absorption (7). Clarithromycin has been shown to inhibit gastrointestinal CYP3A4 and thereby increase the absorption of midazolam, a substrate of CYP3A4 but not of P-gp (6). Clarithromycin has also been shown to increase the absorption of digoxin, a substrate of P-gp but not of CYP3A4 (24). Since all of the HIV-1 protease inhibitors are substrates of CYP3A4 (5) and are transported by P-gp (12, 17, 25), the increase in the AUC for amprenavir following clarithromycin pretreatment could be due to one or both of these mecha-

nisms. There was a near-significant (P = 0.065) negative relationship between the baseline amprenavir AUC and the magnitude of the increase in the AUC following clarithromycin pretreatment. This suggests that those subjects with a low baseline amprenavir AUC, possibly resulting from greater first-pass clearance mediated by CYP3A4 and/or P-gp, have a larger
interaction with clarithromycin, since it interferes with those processes that act to reduce absorption. Similar mechanisms explain the effects when two protease inhibitors are given together, as when ritonavir is given with either saquinavir (9) or indinavir (10). Modeling of these interactions suggests that the main effect of ritonavir on indinavir is a reduction in systemic clearance via inhibition of hepatic CYP3A4 metabolism (10), whereas the effect of ritonavir on saquinavir is mediated mainly through a reduction in first-pass gastrointestinal CYP3A4 metabolism (9). It is not yet feasible to quantify the relative contribution of P-gp versus CYP3A4 to these interactions in vivo. Irrespective of the mechanisms for these interactions, these data indicate that clarithromycin and amprenavir can be given together with no need for dosage adjustment.

ACKNOWLEDGMENTS

This study was supported by a grant from Glaxo Wellcome, Inc. Appreciation is expressed to Cindy Rawls (Glaxo Wellcome, Inc.), who performed the amprenavir assays; to Clark March (School of Pharmacy, VCU), who performed the ERMIB scintillation counts; and to the nurses and staff of the Center for Drug Studies at the Virginia Commonwealth University School of Pharmacy.

REFERENCES