ANTIMICROBIAL AGENTS AND CHEMOTHERAPY, Nov. 1997, p. 2428–2432 Vol. 41, No. 11
0066-4804/97/$04.00+0

Pharmacokinetics of Saquinavir, Zidovudine, and Zalcitabine in Combination Therapy

GEERTRUI F. VANHOVE,[1] HELEN KASTRISSIOS,[2] JEAN-MICHEL GRIES,[2] DAVIDE VEROTTA,[2] KYUNGSOO PARK,[2] ANN C. COLLIER,[3] KATHLEEN SQUIRES,[4] LEWIS B. SHEINER,[2] AND TERRENCE F. BLASCHKE[1]*

Division of Clinical Pharmacology, Stanford University, Stanford,[1] and Department of Laboratory Medicine, University of California San Francisco, San Francisco,[2] California; Division of Infectious Diseases, University of Washington School of Medicine, Seattle, Washington[3]; and Division of Infectious Diseases, University of Alabama at Birmingham, Birmingham, Alabama[4]

Received 20 September 1996/Returned for modification 3 February 1997/Accepted 21 August 1997

We investigated the pharmacokinetics of zidovudine, zalcitabine, and saquinavir in AIDS Clinical Trial Group protocol 229. Patients received either saquinavir, zalcitabine, or a combination of both, together with zidovudine three times a day. Approximately 100 patients were enrolled in each treatment arm, and intensive pharmacokinetic studies were performed on about 25 patients per arm at weeks 1 and 12. We estimated the pharmacokinetic parameters of all three drugs by using parametric and nonparametric methods. The mean values of the pharmacokinetic parameters of zidovudine (clearance [CL]/bioavailability [*F*], 168 liters/h; volume of distribution [*V*]/*F*, 185 liters; half-life, 0.76 h) and zalcitabine (CL/*F*, 25 liters/h; *V*/*F*, 92.2 liters; half-life, 2.7 h) were similar to those reported previously. For saquinavir, the mean pharmacokinetic parameter estimates using parametric methods were as follows: maximum concentration of drug in serum [C_{max}], 70.8 ng/ml; time to C_{max}, 3.11 h; area under the curve, 809 ng · h/ml; CL/*F*, 989 liters/h; *V*/*F*, 1,503 liters; half-life, 1.38 h. For all three drugs, clearance decreased with age. Weight did not influence the clearance of zidovudine, but the clearance of zalcitabine and saquinavir increased with weight. There were no differences in pharmacokinetic parameters between study weeks and arms, suggesting that there is no change in kinetics with chronic administration and that there are no significant pharmacokinetic interactions among these three drugs.

Human immunodeficiency virus protease inhibitors, such as saquinavir, are a promising new class of antiretroviral drugs. AIDS Clinical Trial Group (ACTG) protocol 229 was designed to test the hypothesis that a combination of saquinavir, zidovudine, and zalcitabine would be more effective than the combination of saquinavir with a nucleoside analog or the combination of two nucleoside analogs. The primary results of this study have been presented elsewhere (3).

Individualization of drug therapy requires an understanding of the subject-specific pharmacokinetics of a drug, as well as of the relationship between drug concentrations (drug exposure) and biological effects. One of the sources of variability in the individual response to similar or identical doses of antiretroviral therapy observed in clinical studies may be inter- and intrapatient variabilities in the pharmacokinetics of prescribed drugs. While zidovudine and zalcitabine pharmacokinetics have been described previously (4, 5), the magnitude of the variability in the pharmacokinetics of saquinavir during chronic administration has not been reported earlier. In this paper, we present standard nonparametric and parametric analyses of the pharmacokinetics of zidovudine and zalcitabine, and for saquinavir we present an additional nonparametric analysis describing the pharmacokinetic data with longitudinal splines (12). The accompanying paper (11a) reports on other sources of variability in individual response and investigates the relationship between exposure to these drugs and viral and immunologic responses.

* Corresponding author. Mailing address: Division of Clinical Pharmacology, Room S-025, Stanford University Medical Center, Stanford, CA 94305-5130. Phone: (415) 725-4632. Fax: (415) 725-8020.

MATERIALS AND METHODS

Study design. ACTG 229 was a phase II randomized study of three treatment regimens. The sample size was approximately 100 patients per treatment arm. Patients in arm 1 received saquinavir at 600 mg three times a day (TID) (Invirase [formerly Roche 31-8959]; Hoffman-La Roche, Nutley, N.J.) plus open-label zidovudine at 200 mg TID (Retrovir; Burroughs Wellcome Company, Research Triangle Park, N.C.). Patients in arm 2 received saquinavir 600 at mg TID plus zalcitabine at 0.75 mg TID (Hivid; Hoffman-La Roche) plus open-label zidovudine at 200 mg TID. Patients in arm 3 received zalcitabine at 0.75 mg TID plus open-label zidovudine at 200 mg TID. The initial treatment duration was 24 weeks, but all of the patients were offered continuation of the same blinded regimen for up to 56 weeks until a common closing date. Patients were required to have documented HIV infection, $CD4^+$ cell counts between 50 and 301 cells/mm^3 within 30 days of study entry, and at least 4 months of prior zidovudine therapy with no toxicity at 600 mg/day. For further details regarding design and patient eligibility criteria, see reference 3.

Pharmacokinetic study design, sampling, and analytical methods. In a subgroup of patients at three designated sites, intensive blood sampling over one dosing interval was performed at week 1 and again at week 12, yielding intensive pharmacokinetic studies of 20 to 25 patients per arm. Samples were collected immediately predose and at 1, 2, 3, 4, 6, and 8 h after dosing. A breakfast was consumed 30 min before taking the dose, since it had been shown that the presence of food increases the bioavailability (*F*) of saquinavir. All study medications were taken simultaneously, and patients did not receive additional study medications within the pharmacokinetic assessment sampling period.

Plasma was separated by centrifugation within half an hour of blood collection, stored at −20°C, and shipped. Samples were assayed for saquinavir by a validated radioimmunoassay developed by Roche within the Bioanalytical Group of the Department of Metabolism and Pharmacokinetics, Huntingdon Center Ltd., Cambridgeshire, United Kingdom. Briefly, standards and samples were incubated overnight at ambient temperature with sheep saquinavir antiserum and iodinated tracer (^{125}I-tyrosyl-saquinavir). Bound and free saquinavir were separated by addition of a second antibody covalently coupled to cellulose followed by an incubation (20 min) and a centrifugation step. Radioactivity was measured in the bound fraction using a gamma counter. The detection limit was 1 ng/ml. The mean levels of interassay precision for standards and quality control samples were ±8.0% and ±9.4%, respectively. The mean levels of intra-assay precision and accuracy for the quality control samples were ±5.0% and +2.4%.

Zidovudine was assayed by high-pressure liquid chromatography with UV detection. The detection limit was 25 ng/ml. The mean levels of interassay precision for standards and quality control samples were ±3.8% and ±4.4%,

TABLE 1. Mean pharmacokinetic parameters of zidovudine and zalcitabine for all arms at weeks 1 and 12[a]

Drug (no. of patients) and method	Log phase (h)	k_a (h^{-1})	C_{max} (ng/ml)	T_{max} (h)	AUC (ng · h/ml)	CL/*F* (liters/h)	*V*/*F* (liters)	$t_{1/2}$ (h)
Zidovudine (73)								
Nonparametric		NA[b]	435 ± 23.8	2.12 ± 0.09	1,020 ± 44.1	243 ± 15.0	NA	NA
Parametric		0.52	347 ± 14.7	1.29 ± 0.06	1,260 ± 36.1	168 ± 4.43	185 ± 28.0	0.76 ± 0.11
Zalcitabine (53)								
Nonparametric		NA	6.97 ± 0.26	2.69 ± 0.14	29.5 ± 1.07	27.5 ± 1.07	NA	NA
Parametric	0.649	1.48 ± 0.19	5.31 ± 0.16	1.75 ± 0.10	31.8 ± 1.15	25.0 ± 0.79	92.2 ± 1.93	2.70 ± 0.12

[a] The values are means ± SEM calculated as described in Materials and Methods.
[b] NA, not available.

respectively. The mean levels of intra-assay precision and accuracy for the quality control samples were ±7.3% and +5.7%.

Zalcitabine was assayed by using solid-phase extraction coupled to atmospheric pressure ionization and mass spectrometry/mass spectrometry detection. The detection limit was 0.5 ng/ml. The mean levels of interassay precision for standards and quality control samples were ±6.5% and ±8.0%, respectively. The mean levels of intraassay precision and accuracy for the quality control samples were ±4.8% and +0.27%.

Pharmacokinetic analysis. For patients in the intensive pharmacokinetics group, we estimated C_{max}, T_{max}, AUC, and clearance (CL)/*F* for all three drugs, for each week and arm separately, by standard nonparametric methods. C_{max} was established as the maximum observed concentration of drug in plasma and T_{max} was as the time to C_{max}. The area under the plasma concentration-time curve (AUC) was calculated by using the trapezoidal rule. CL/*F* was calculated as dose/AUC. Mean values were calculated as follows. For patients who had intensive pharmacokinetic sampling done at weeks 1 and 12, the mean of the parameter estimates for both weeks was used as the mean value. For patients who had intensive pharmacokinetic sampling at only 1 week, the single estimate was used as the mean value. Subsequently, the arithmetic average, standard deviation (SD), and standard error of the mean (SEM) of all of the individual means were calculated. The interpatient variability, expressed as the percent coefficient of variation (CV), was calculated by dividing the SD of each parameter estimate by its mean and multiplying the result by 100. The intrapatient-interoccasion variability was obtained by calculating the CV of the ratio of individual week 12 to week 1 parameter estimates for C_{max}, T_{max}, AUC, and CL/*F*.

In addition, the intensive pharmacokinetic data for all three drugs were also fitted to a one-compartment model with first-order absorption by using the NONMEM software program (1). This program produces estimates of the population mean parameters of the one-compartment model, including lag phase, absorption rate constant (k_a), CL/*F*, and volume of distribution V/*F*. The observations at weeks 1 and 12 were modeled together, as statistical analysis of the estimates obtained by the nonparametric methods described above showed no difference in parameters between weeks. We investigated the influences of age, weight, and length of prior zidovudine therapy on the individual CL and *V* of all three drugs. The interindividual variability in all parameters was assumed to follow a log-normal distribution. For zidovudine, we did not include any interindividual variability for k_a because of limited data during the absorption phase. Intraindividual variability was modeled by using a constant plus proportional variance model according to the expression $C_{obs} = C_e + \sqrt{(C_e^2 + \theta^2)} \cdot \varepsilon_1$, in which C_{obs} is the observed concentration, C_e is the expected concentration, θ is a parameter to be estimated, and ε_1 is a normally distributed random variable with mean zero and unknown variance σ^2. The population estimates of the intensively studied group provided a prior distribution from which we produced individual empirical Bayes estimates of each patient's parameters. We calculated the individual estimates of C_{max}, T_{max}, AUC, and half-life ($t_{1/2}$) by using the individual empirical Bayes estimates for k_a, CL/*F* and *V*/*F* and calculated the mean, SD, and SEM of the different parameters from the individual Bayes estimates. Interpatient variability, expressed as the percent CV, was subsequently calculated by dividing the standard deviation of each parameter by its mean and multiplying the result by 100.

Since the saquinavir data were not well described by any standard pharmacokinetic model, we used longitudinal splines (12) to describe the individual saquinavir concentration-versus-time profiles, from which individual C_{max}, T_{max}, AUC, and CL/*F* values could then be estimated. A longitudinal spline is composed of a template spline, common to all subjects, and a distortion spline describing the subject's difference from the template. The individual longitudinal spline estimates are empirical Bayes estimates, as above. Each individual spline was constrained to have identical values at the beginning and end of a (steady-state) dosing interval, to have a decreasing tail, and to have its T_{max} near the population mode of individual data-based T_{max} values. The mean, SD, SEM, and interpatient and intrapatient-interoccasion variabilities were computed as described above for the standard nonparametric methods.

Statistical analysis. Analysis of variance was used to test for differences in weight, age, and prior zidovudine therapy among treatment arms. Analysis of variance was also used to test for differences among treatment arms in the estimates of C_{max}, T_{max}, AUC, and CL/*F* of zidovudine obtained by nonparametric methods and in the individual empirical Bayes estimates of k_a, CL/*F*, *V*/*F*, C_{max}, T_{max}, AUC, and $t_{1/2}$ of zidovudine obtained by parametric methods. We used a paired *t* test to test for differences in pharmacokinetic parameters obtained by nonparametric methods for all three drugs between weeks. In doing so, we pooled individual drug kinetics across treatment arms. An unpaired *t* test was used to test for differences between treatment arms in the estimates of C_{max}, T_{max}, AUC, and CL/*F* obtained by nonparametric methods and in the individual empirical Bayes estimates of k_a, CL/*F*, *V*/*F*, C_{max}, T_{max}, AUC, and $t_{1/2}$ obtained by parametric methods for saquinavir and zalcitabine. For all estimates obtained by nonparametric methods, we used the mean values as described above for pharmacokinetic analysis to test for differences between treatment arms.

RESULTS

There were no significant differences in weight, age, or prior zidovudine therapy among treatment arms for patients in the intensive pharmacokinetics groups. The mean ages (±SD) of patients exposed to saquinavir, zidovudine, and zalcitabine were 41.5 ± 9.74, 39.4 ± 9.66, and 38.8 ± 9.21 years, respectively. The mean weights (±SD) of patients exposed to saquinavir, zidovudine, and zalcitabine were 78.1 ± 11.6, 77.1 ± 11, and 77.2 ± 11.1 kg, respectively, and the median duration of prior zidovudine therapy (range) was 740 days (15 to 2,252 days).

Zidovudine and zalcitabine. Table 1 shows the results expressed as mean values ± SEM from parametric and nonparametric analyses of the pharmacokinetics of zidovudine and zalcitabine. The mean values based on parametric or nonparametric individual estimates were comparable. In the parametric analysis of the zidovudine population, CL/*F* appeared to decrease nonlinearly with age and was modeled by using the equation $CL = \theta_1 \cdot \exp[\theta_2 \cdot (age - 50)]$ for ages >50 and θ_1 for ages ≤50, where θ_1 and θ_2 were population values to be estimated and 50 was the age estimated by the NONMEM program at which clearance started to decrease significantly with age. Of 73 patients exposed to zidovudine, six were older than 50.

In the zalcitabine population analyses, CL appeared to diminish with age and to increase with weight. *V* was not influenced by these covariates. CL was modeled by using the equation $CL = \theta_1 \cdot \exp[\theta_2 \cdot (age - mean\ age)] + \theta_3 \cdot weight$, where θ_1, θ_2, and θ_3 were population values to be estimated. We also included a nonrandom lag phase to improve the fit.

Scatterplots of the data from the intensively studied group and the fitted curves and concentration-versus-time curves of individual representative patients are shown in Fig. 1. As is apparent in the right-hand panels, the pharmacokinetics of zidovudine and zalcitabine are reasonably well described by a one-compartment model. In a number of cases, however, the

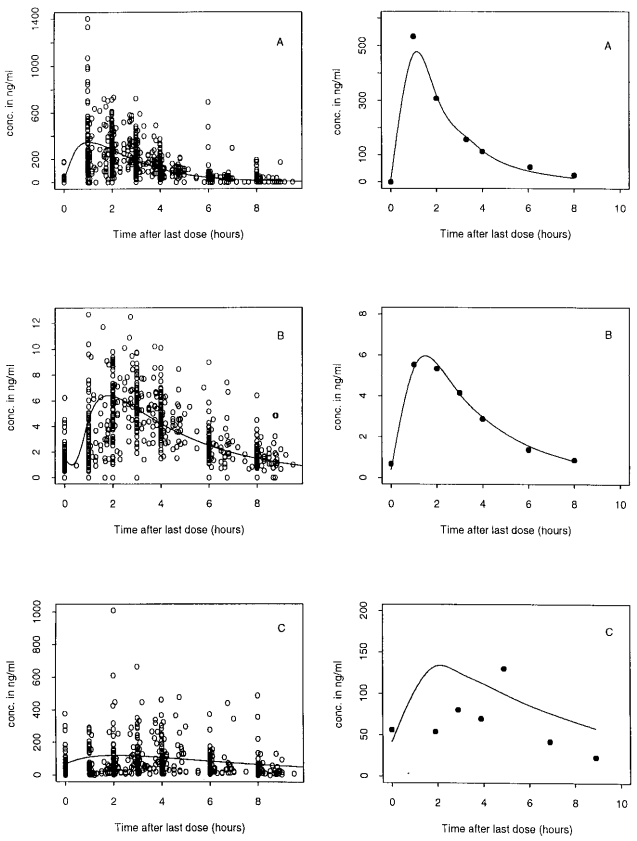

FIG. 1. Concentration-versus-time curves of patients in the intensive group. (Left) The concentrations (conc.) of zidovudine (A), zalcitabine (B), and saquinavir are plotted versus time after dose administration to intensive-pharmacokinetics patients. The solid lines represent the population mean estimated from the NONMEM fit, using a one-compartment model (as described in Materials and Methods). (Right) The observed concentrations (closed circles) of zidovudine (A), zalcitabine (B), and saquinavir are plotted versus time after dose administration for a representative individual patient. The solid lines are predicted values (Bayes estimates generated by NONMEM).

observed concentrations showed considerable departure from the smooth model predictions, resulting in different estimates of C_{max} and T_{max} by parametric and nonparametric methods.

There were no significant differences between weeks or treatment arms in the individual estimates of k_a, CL/*F*, *V*/*F*, C_{max}, T_{max}, AUC, or $t_{1/2}$ of zidovudine and zalcitabine obtained by parametric or nonparametric methods (data not shown).

Table 2 shows the interpatient and intrapatient-interoccasion variabilities of the different parameters. For both drugs, but for zidovudine in particular, the interpatient and intrapatient-interoccasion variabilities were fairly large.

Saquinavir. Table 3 shows the results expressed as means ± SEM for parametric and nonparametric analyses of the pharmacokinetics of saquinavir. We chose to present all three methods to allow comparison to previous or future analyses of saquinavir pharmacokinetics using one of the three methods. As can be seen in Table 3, the parameter values estimated by parametric or nonparametric methods were fairly similar, except for C_{max}, for which the estimate was much lower by parametric methods. This is probably due to the fact that the data are not well described by the pharmacokinetic model used here. A scatterplot of the data of the intensively studied group and the fitted curve is shown in Fig. 1 together with a concentration-versus-time curve of an individual patient. CL appeared to diminish with age and to increase with weight. *V* was not influenced by these covariates. CL was modeled by using the equation $CL = \theta_1 \cdot \exp[\theta_2 \cdot (\text{age} - \text{mean age})] + \theta_3 \cdot \text{weight}$, where θ_1, θ_2, and θ_3 were population values to be estimated. As demonstrated in Fig. 1C, the plasma concentration-versus-time curve for saquinavir showed considerable variability and was not well described by the one-compartment pharmacokinetic model. For this reason, we have also described the data by means of splines (see Materials and Methods).

Table 4 shows the interpatient and intrapatient-interoccasion variabilities of the different parameters of saquinavir obtained by different methods. For all of the methods used, the interpatient and intrapatient-interoccasion variabilities were very large. This is further illustrated in Fig. 2, which provides an overview of the spread of the estimates of C_{max}, AUC, and CL/*F*. For the estimates per week, only the estimates based on nonparametric methods are shown since observations at weeks 1 and 12 were modeled together by using parametric methods. As can be seen in Fig. 2, the spread of the estimates of CL/*F* was less when longitudinal splines or parametric methods were used. There were no significant differences between weeks or treatment arms in the individual empirical Bayes estimates of k_a, CL/*F*, *V*/*F*, C_{max}, T_{max}, AUC, and $t_{1/2}$ obtained by parametric methods, standard nonparametric methods, or nonparametric spline methods (data not shown).

TABLE 2. Interpatient and intrapatient-interoccasion variabilities of zidovudine and zalcitabine pharmacokinetic parameters

Analysis method, variability, and drug	CV (%)[a]						
	k_a	C_{max}	T_{max}	AUC	CL/*F*	*V*/*F*	$t_{1/2}$
Nonparametric							
Interpatient							
Zidovudine	NA[b]	47	36	37	53	NA	NA
Zalcitabine	NA	27	38	26	28	NA	NA
Intrapatient-interoccasion							
Zidovudine	NA	75	58	85	67	NA	NA
Zalcitabine	NA	27	51	20	24	NA	NA
Parametric, interpatient							
Zidovudine	NA	36	43	27	23	130	120
Zalcitabine	92	22	41	26	23	15	32

[a] The values were determined as described in Materials and Methods.
[b] NA, not available.

TABLE 3. Pharmacokinetic parameters of sequinavir for all arms at weeks 1 and 12[a]

Analysis method (no. of patients)	k_a (h)	C_{max} (ng/ml)	T_{max} (h)	AUC (ng · h/ml)	CL/*F* (liters/h)	*V*/*F* (liters)	$t_{1/2}$ (h)
Nonparametric							
Standard (46)	NA[b]	185 ± 24.2	3.98 ± 0.2	621 ± 68.8	1,740 ± 190	NA	NA
Spline (45)	NA	163 ± 23.2	3.76 ± 0.09	717 ± 65.2	1,110 ± 74.5	NA	NA
Parametric (45)	0.137	70.8 ± 6.14	3.11 ± 0.27	809 ± 70.2	989 ± 77.0	1,503 ± 188	1.38 ± 0.24

[a] The values shown are means ± SEM calculated as described in Materials and Methods.
[b] NA, not available.

DISCUSSION

As presented in Results, both interpatient and intrapatient-interoccasion variabilities in the kinetics and exposure of saquinavir, zidovudine, and zalcitabine are large. It has previously been shown that the *F* of this formulation of saquinavir is about 18 times higher in the presence of food, but even with food the extent of absorption is only 4% with a CV of 60% (14). The low *F* is thought to be due largely to first-pass metabolism by cytochrome P-450 CYP3A4. It is well known that small changes in the extent of absorption or metabolism of drugs with extensive first-pass metabolism can result in large changes and substantial variability in the concentrations in plasma. The *F* of zidovudine is about 64% in the absence of food. The incomplete absorption of zidovudine is the result of first-pass hepatic glucuronidation (2). The administration of a standard breakfast has been shown to prolong T_{max} and decrease C_{max} without any change in the total AUC or CL of zidovudine (9, 11). Recent studies in patients have also demonstrated considerable between-patient variability in zidovudine *F*, with low *F* being associated with the presence of mild diarrhea and/or low $CD4^+$ counts (6). These factors may explain the high interpatient and intrapatient-interoccasion variabilities of zidovudine observed in this study.

The values obtained for the pharmacokinetic parameters of saquinavir by using splines are generally intermediate between the standard nonparametric methods obtained without splines and the parametric model estimates. However, the CL/*F* obtained by using splines is about two-thirds of that obtained without splines and is comparable to the value estimated by NONMEM using the one-compartment pharmacokinetic model. Although the mean AUC increases by only 17% using splines, the mean CL/*F* calculated as dose/AUC decreases by 33%. This appears to be a result of the distribution of the AUC. The fact that the parametric model and the nonparametric spline method generate values that are not too dissimilar is reassuring evidence that these estimates are probably close to the true population values.

For the other two drugs, the CL/*F* values obtained by nonparametric methods are comparable to those obtained by using a parametric model. Moreover, the mean values of the pharmacokinetic parameters of zidovudine are similar to those reported in previous studies, taking into account the 64% bioavailability of zidovudine and a mean population weight of 70 kg (5; for a review, see reference 13). The finding that the CL of zidovudine decreases in patients over 50 is consistent with the finding of a longer $t_{1/2}$ in patients over 60 (7). For zalcitabine, the values of the pharmacokinetic parameters reported here are similar to those reported previously (4). There have been no previous reports on the influence of age on the pharmacokinetics of zalcitabine with which to compare our finding that CL decreases with age.

The absence of differences in the pharmacokinetic parameters of zidovudine and zalcitabine between arms indicates that these drugs do not affect each other's pharmacokinetics. This agrees with previous findings (8). No differences in pharmacokinetic parameters for saquinavir between arms were observed. However, power calculations demonstrate that there was only a 30% chance of finding as much as a 30% difference in the AUC between arms 1 and 2, indicating that only interactions that result in large effects could have been detected.

As mentioned in the introduction, understanding of the pharmacokinetics of a drug is a prerequisite for optimization and individualization of drug therapy. Although high interpatient variability in pharmacokinetics generally implies that therapeutic drug monitoring may be useful in detecting patients with undetectable or very low drug concentrations, additional high intrapatient-interoccasion variability decreases the utility of such drug monitoring. In a recently published monotherapy study, exposure to a saquinavir dose of 3,600 mg/day was associated with a higher mutational rate and de-

TABLE 4. Interpatient and intrapatient-interoccasion variabilities of the pharmacokinetic parameters of saquinavir

Variability and analysis method (no. of patients)	CV (%)[a]					
	C_{max}	T_{max}	AUC	CL/*F*	*V*/*F*	$t_{1/2}$
Interpatient						
Parametric (45)	58	58	58	52	84	120
Nonparametric						
Standard (46)	89	34	75	74	NA[b]	NA
Spline (45)	95	17	61	45	NA	NA
Intrapatient-interoccasion, nonparametric						
Standard (41)	105	73	88	62	NA	NA
Spline (42)	67	37	61	47	NA	NA

[a] The data shown were calculated as described in Materials and Methods.
[b] NA, not available.

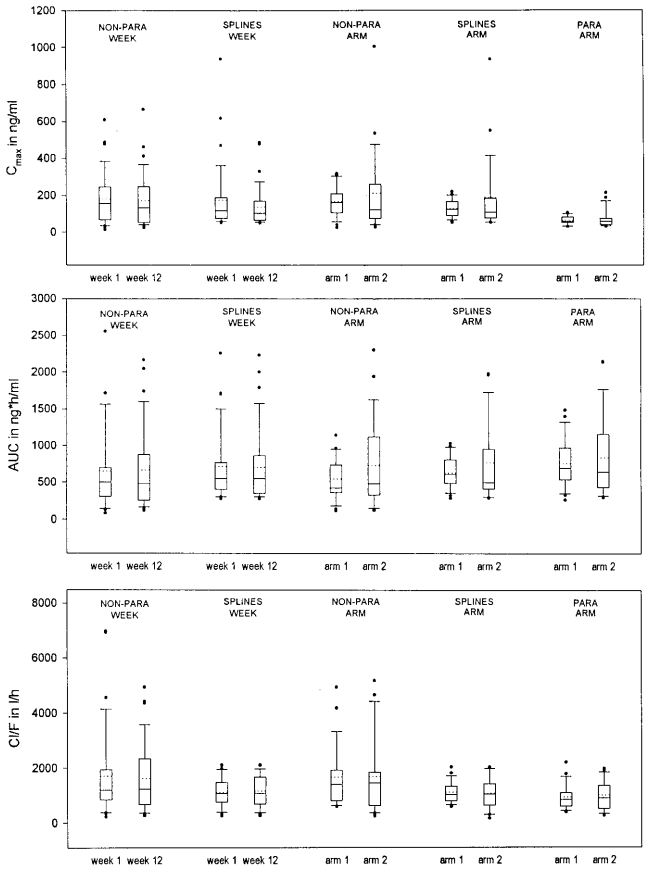

FIG. 2. C_{max}, AUC, and CL/*F* for saquinavir estimated by parametric and nonparametric methods. Box plots of estimates of C_{max}, AUC, and CL/*F* for saquinavir. Abbreviations: NON-PARA, estimates obtained by using standard nonparametric methods, SPLINES, estimates obtained by using longitudinal splines, PARA, estimates obtained by using parametric methods. The interior line in each box is located at the median of the data. The dotted line represents the mean of the data. Each box encompasses the 25th through the 75th percentiles. The whiskers represent the 10th and 90th percentiles, and the dots represent outliers.

velopment of drug resistance than exposure to a dose of 7,200 mg/day (10), stressing the importance of high drug exposure. Unfortunately, the high intrapatient-interoccasion pharmacokinetic variability of this formulation of saquinavir suggests that drug monitoring on one occasion would not be predictive of other occasions. Since we were able to demonstrate a positive relationship between exposure to saquinavir, as part of a triple combination, and a maximum increase in $CD4^+$ cell count and a maximum decrease in the level of RNA in plasma in the study described in the following paper (11a), therapeutic drug monitoring may be justifiable if a new formulation of saquinavir with a larger *F* becomes available for patients in whom the recommended doses of saquinavir result in low immunological and viral responses.

ACKNOWLEDGMENTS

This study was supported by grants AI 27666, AI 27663, GM 26691, AI 27664, M01-RR00037, and M01-RR00070; the Collen Research Foundation, and the ACTG.

We thank R. Bassett and D. Schoenfeld for assistance with the data; A. Brown, F. Stewart, and P. Williams for providing us with the assay results, and the nurses and staff of the University of Alabama, Stanford University, and the University of Washington, Seattle, for carrying out the intensive pharmacokinetic studies.

REFERENCES

1. **Beal, S. L., and L. B. Sheiner.** 1991. NONMEM user's guide. NONMEM Project Group, University of California at San Francisco, San Francisco, Calif.
2. **Blum, M. R., S. H. T. Liao, S. S. Good, and P. de Miranda.** 1988. Pharmacokinetics and bioavailability of zidovudine in humans. Am. J. Med. **85**(Suppl. 2A)**:**189–194.
3. **Collier, A. C., R. W. Coombs, D. A. Schoenfeld, R. Bassett, et al.** 1996. Treatment of human immunodeficiency virus infection with saquinavir, zidovudine and zalcitabine. N. Engl. J. Med. **334:**1011–1017.
4. **Klecker, R. W., J. M. Collins, R. C. Yarchoan, R. Thomas, N. McAtee, S. Broder, and C. E. Myers.** 1988. Pharmacokinetics of 2′,3′-dideoxycytidine in patients with AIDS and related disorders. J. Clin. Pharmacol. **28:**837–842.
5. **Klecker, R. W., J. M. Collins, R. Yarchoan, R. Thomas, J. F. Jenkins, et al.** 1987. Plasma and cerebrospinal fluid pharmacokinetics of 3′-azido-3′ deoxythymidine: a novel pyrimidine analog with potential application for the treatment of patients with AIDS and releated diseases. Clin. Pharmacol. Ther. **41:**407–412.
6. **MacNab, K. A., J. Gill, L. R. Sutherland, N. De Boer Visser, and V. Church.** 1993. Erratic zidovudine bioavailability in HIV seropositive patients. J. Antimicrob. Chemother. **31:**421–428.
7. **Matthews, S. J., R. J. Cersosimo, and M. L. Spivack.** 1991. Zidovudine and other reverse transcriptase inhibitors in the management of human immunodeficiency virus-related disease. Pharmacotherapy **11:**419–449.
8. **Meng, T. C., M. A. Fischl, A. M. Boota, et al.** 1992. Combination therapy with zidovudine and dideoxycytidine in patients with advanced human immunodeficiency virus infection. Ann. Intern. Med. **116:**13–20.
9. **Ruhnke, M., F. E. Bauer, M. Seifert, M. Trautmann, H. Hille, and P. Koeppe.** 1993. Effects of standard breakfast on pharmacokinetics of oral zidovudine of patients with AIDS. Antimicrob. Agents Chemother. **3:**2153–2158.
10. **Schapiro, J. M., M. A. Winters, F. Stewart, B. Efron, J. Norris, M. J. Kozal, and T. C. Merigan.** 1996. The effect of high dose saquinavir on viral load and $CD4^+$ T-cell counts in HIV-infected patients. Ann. Intern. Med. **124:**1039–1050.
11. **Unadkat, J. D., A. C. Collier, S. S. Crosby, D. Cummings, K. E. Opheim, and L. Corey.** 1990. Pharmacokinetics of oral zidovudine (azidothymidine) in patients with AIDS when administered with and without a high fat meal. AIDS **4:**229–232.

11a. **Vanhove, G. F., J.-M. Gries, D. Verotta, L. B. Sheiner, R. Coombs, A. C. Collier, and T. F. Blaschke.** 1997. Exposure-response relationships for sequinavir, zidovudine, and zalcitabine in combination therapy. Antimicrob. Agents Chemother. **41:**2433–2438.

12. **Verotta, D.** 1995. New approaches to self-modeling non-linear regression. *In* D. Z. D'Argenio (ed.), Advanced methods of pharmacokinetic and pharmacodynamic system analysis II. Plenum Press, New York, N.Y.
13. **Wilde, M. I., and H. D. Langtry.** 1993. Zidovudine. An update of its pharmacodynamic and pharmacokinetic properties and therapeutic efficacy. Drugs **46:**515–578.
14. **Williams, P. E. O., G. J. Muirhead, M. J. Madigen, A M. Mitchel, and T. Shaw.** 1992. Disposition and bioavailability of the protease inhibitor, Ro 31-8959, after single doses in healthy volunteers. Br. J. Clin. Pharmacol. **34:**155P–156P.