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Pyrimethamine Pharmacokinetics in Human Immunodeficiency Virus-Positive Patients Seropositive for *Toxoplasma gondii*

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Pyrimethamine pharmacokinetics were studied in 11 human immunodeficiency virus (HIV)-positive patients who were seropositive for exposure to *Toxoplasma gondii* and were taking zidovudine (AIDS Clinical Trials Group Protocol 102). Pyrimethamine was administered at 50 mg daily for 3 weeks to achieve steady state, and pharmacokinetic profiles were determined after administration of the last dose. Noncompartmental and compartmental analyses were performed. Population pharmacokinetic analysis assuming a one-compartment model yielded the following estimates: area under the 24-h concentration-time curve, 42.7 ± 12.3 μg · h/ml; half-life, 139 ± 34 h; clearance, 1.28 ± 0.41 liters/h; volume of distribution, 246 ± 64 l; and absorption rate constant, 1.5 ± 1.3 liters/h. These values are similar to those seen in subjects without HIV infection. Pyrimethamine pharmacokinetics did not differ significantly in those subjects who were intravenous drug users. Adverse effects were noted in 73% of those initially enrolled in this study, leading to discontinuation for 38%. No association was noted between pyrimethamine levels and the incidence of adverse events. No significant differences were seen in zidovudine pharmacokinetic parameters obtained from studies performed before and during treatment with pyrimethamine. In summary, pyrimethamine exhibited pharmacokinetics in HIV-infected patients that were similar to those in non-HIV-infected subjects and it did not alter the pharmacokinetics of zidovudine in these patients.

Toxoplasma encephalitis is the most common opportunistic infection causing focal brain disease in patients with AIDS (13, 16). The incidence of toxoplasmic encephalitis is directly proportional to the prevalence of antibodies to *Toxoplasma gondii* in any given population (15). Over 95% of toxoplasmic encephalitis cases are due to reactivation of a chronic (latent) infection, usually when the CD4 lymphocyte count falls below 200/mm^3. In the United States, 10 to 40% of adults with AIDS are latently infected and it is estimated that 30% of these patients will develop toxoplasmic encephalitis (7, 14).

Pyrimethamine [2,4-diamino-5-(*p*-chlorophenyl)-6-ethylpyrimidine] in combination with a sulfonamide is the standard therapeutic regimen for toxoplasmic encephalitis (12). Oral folinic acid is added to preclude the hematologic toxicity associated with pyrimethamine. A clinical response with this combination has been reported in 68 to 95% of patients with toxoplasmic encephalitis (8, 16). Unfortunately, high toxicity rates have led to the discontinuation of therapy in up to 40% of patients (8, 14). The adverse effects of pyrimethamine are commonly dose-related cytopenias and rash, whereas the adverse effects of sulfonamides include rash, nausea, cytopenia, and nephrotoxicity, including crystalluria, hematuria, radiolucent stones, interstitial nephritis, and renal failure. The combination of oral pyrimethamine and oral or parenteral clindamycin seems comparable in efficacy to pyrimethamine plus sulfonamides (4, 9, 14) and is the recommended alternative regimen for a sulfa-intolerant patient. Again, the incidence of adverse effects, including nausea, vomiting, diarrhea, rash, neutropenia, and pseudomembranous colitis, is high. Alternative therapeutic options are limited. To date, there is no information correlating serum drug concentrations with toxicities.

An understanding of the pharmacokinetics of pyrimethamine in patients with AIDS could be helpful in developing treatment regimens that minimize the side effects but maintain efficacy. Little information is currently available. Two studies of patients with AIDS being treated for toxoplasma encephalitis suggested that the half-lives of pyrimethamine in serum were much shorter than those typically found in noninfected individuals. In one study, half-lives of 22.7 and 36.2 h were found in the two patients fully evaluated (24). In the other, the peak and trough values reported allowed the calculation of half-lives ranging from 33 to 84 h (11). Half-lives in healthy human immunodeficiency virus (HIV)-negative patients have ranged from 35 to 175 h (24), with reported averages of 83 (2) and 111 (18) h. In addition, little is known of the drug interaction between pyrimethamine and zidovudine, the mainstay of HIV treatment.

We undertook an open-label, steady-state pharmacokinetic study of pyrimethamine in HIV-infected patients seropositive for exposure to *T. gondii*. We used the standard 50-mg daily dose recommended for treating toxoplasma encephalitis. In addition, we studied the effects of pyrimethamine on zidovudine pharmacokinetics.

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MATERIALS AND METHODS

Subjects. Twenty-six patients were enrolled in the National Institute of Allergy and Infectious Diseases AIDS Clinical Trials Group Protocol 102. Patients were enrolled until 16 had completed the study treatment period (8 homosexual males and 8 intravenous drug users; 13 from the Bronx Veterans' Affairs Medical Center and 3 from the University of North Carolina).

Eligible patients had laboratory evidence of HIV infection (positive antibody enzyme-linked immunosorbent assay and Western blot [immunoblot], p24 antigen, or culture) with an absolute CD4 lymphocyte count of $\leq$500/mm^3 and serological evidence of exposure to *T. gondii* (Sabin-Feldman dye, whole-cell agglutination, or immunoglobulin G immunofluorescence test) but no clinical manifestations of active toxoplasmic infection. They had been on a stable regimen of zidovudine ($\geq$500 mg/day) for at least 4 weeks and were continued on this dosage for the duration of this study. All subjects were at least 18 years of age and had a Karnofsky performance score of $\geq$60, total bilirubin of $\leq$1.5 mg/100 ml, serum aspartate aminotransferase activity of $\leq$3 times the upper limit of the normal range, serum creatinine levels of $<$1.5 times the upper limit of the normal range (or a calculated creatinine clearance of $\geq$50 ml/min for patients with "wasting" syndrome), $<$3+ proteinuria, an absolute neutrophil count of $>$1,500/mm^3, and hemoglobin levels of $\geq$10.0 g/dl.

Patients were excluded if they had a history of prior toxoplasmic encephalitis or of sensitivity to the study medications, had evidence of another serious opportunistic infection or central nervous system impairment, were unable to take oral medication reliably, had a malabsorption syndrome, or were pregnant or lactating. Patients were on a variety of medications during this study. However, treatment with macrolides, sulfonamides, phenytoin, dapsone, immunomodulators, amphotericin B, ganciclovir, probenecid, benzodiazepines, cytotoxic chemotherapy, antifolates, other antiretroviral agents, and investigational agents was forbidden within the 14 days prior to study entry and during this study. An exception was made for patient 8, who was started on erythromycin on day 14.

Study design. All patients received an oral loading dose of 200 mg of pyrimethamine on day 1. Subsequently, patients took 50 mg of pyrimethamine (two 25-mg tablets) and 10 mg of oral folinic acid daily for 21 days, as well as zidovudine in the dose prescribed prior to enrollment. Trough levels of pyrimethamine (drawn immediately before administration of the daily dose) were obtained weekly during this study.

An initial study of zidovudine pharmacokinetics was performed prior to the administration of pyrimethamine on day 1. Serum specimens for drug analysis were obtained immediately before oral intake of zidovudine and at 0.25, 0.5, 0.75, 1.0, 1.5, 2.0, 3.0, and 4.0 h afterwards. The pharmacokinetics of both zidovudine and pyrimethamine were studied after 21 days of pyrimethamine therapy, during which a steady state was achieved. Serum specimens were drawn just prior to a final oral dose of pyrimethamine and at 0.25, 0.50, 0.75, 1.0, 2.0, 3.0, 4.0, 5.0, 6.0, 8.0, 10.0, 12.0, 16.0, 24.0, 36.0, 72.0, and 96.0 h afterwards.

Patient evaluation and follow-up. A complete medical history and physical examination were performed within 1 week prior to study entry. A complete blood cell count with differential, serum chemistries, and a pregnancy test for women of child-bearing potential were performed within 2 weeks prior to entry. At weekly intervals during this study, a brief history, physical examination, complete blood cell count with differential, and serum chemistries were obtained. Patients were hospitalized during the pharmacokinetic evaluations.

Patients who took less than 75% of the study therapy during a 2-week period, as determined by pill count, were considered noncompliant and were discontinued from this study. Patients who missed two consecutive clinic visits without rescheduling were also considered noncompliant and were discontinued.

Toxicity management. Study therapy was discontinued permanently if patients developed an absolute neutrophil count of less than 1,000/mm^3 or a 50% reduction from the baseline count to less than 3,000/mm^3, a platelet count of less than 75,000/mm^3, serum aspartate aminotransferase activity of greater than 5 times the upper limit of normal, a doubling of serum creatinine or an increase to $>$2.0, or any other grade 2 toxicity other than fatigue, headache, or constipation, as defined by the AIDS Clinical Trials Group standard table for grading the severity of adverse experiences.

Blood was drawn for determination of the pyrimethamine concentration at the time of any permanent discontinuation for toxicity. Patients discontinued from study treatment received appropriate medical management. Clinical follow-up and pertinent laboratory evaluations were repeated as needed until there was final resolution or stabilization of toxicity.

Pharmacokinetic analyses. Pyrimethamine concentrations were measured by high-performance liquid chromatography (HPLC). Interday coefficients of variation were 5.3% at 0.2 μg/ml, 1.4% at 0.75 μg/ml, and 2.0% at 2.5 μg/ml; analytical recovery ranged from 97.9 to 103.5% (20). Zidovudine concentrations were measured by ZDV-Trac radioimmunoassay (Incstar Corp., Stillwater, Minn.) using in-house calibrators prepared by supplementing pooled human serum (Scantibodies Laboratory, Santee, Calif.) with known amounts of zidovudine (Sigma Chemical Co., St. Louis, Mo.). Interday coefficients of variation were 7.55% at 139 ng/ml and 7.44% at 1,334 ng/ml; analytical recovery was 101.5% at both concentrations.

Both noncompartmental and compartmental analyses of pyrimethamine concentrations over time were performed. The compartmental analyses provide a complementary interpretation of the data relative to noncompartmental techniques. Zidovudine concentrations over time were analyzed by noncompartmental methods, and the effects of pyrimethamine on the pharmacokinetics of zidovudine were determined by applying a paired Student's *t* test or signed rank test to the difference in the various pharmacokinetic parameters before and after pyrimethamine administration. The signed rank test was preferred in cases in which a Shapiro-Wilk test indicated nonnormality. The interaction of pyrimethamine with zidovudine was also analyzed separately in patients who used intravenous drugs and in nonusers.

Noncompartmental analyses. The terminal elimination rate constant (k_{el}) was estimated in a stepwise log-linear regression beginning with the last three observations. On the basis of this regression fit, a 95% forecast interval was constructed for an observation at the next preceding time. If the actual observed concentration at this time fell within this forecast interval, a new log-linear regression including this observation was performed; if not, the procedure was terminated, and k_{el} was calculated as the negative of the slope of the final line. The forecast interval was the region for which the probability of an observation was 0.95, assuming that the observation arose from the same mean linear relationship used to calculate the interval. Thus, the criterion used corresponded to concluding that there was evidence to suggest a departure from the current line if the concentration observed had a likelihood of less than 5% of having arisen from the current mean linear relationship. The terminal elimination half-life ($t_{1/2}$) was determined as $t_{1/2} = \ln(2)/k_{el}$.

The area under the concentration-time curve from 0 to 24 h (AUC_{0-24}) was calculated by using the trapezoidal rule, and the AUC extrapolated to infinite time ($AUC_{0-\infty}$) was determined by calculating the observed area by the trapezoidal rule and adding the extrapolated area from the last time point to infinity (the final concentration divided by k_{el}). Because no intravenous data were obtained, it was not possible to estimate the bioavailability fraction (F). Thus, oral clearance (CL/F) was calculated as $CL/F = \text{dose}/AUC_{0-24}$, and the apparent volume of distribution (V/F) during the elimination phase was calculated as $V/F = (CL/F)/k_{el}$.

Compartmental analyses. As a complementary adjunct to noncompartmental analyses, serum concentrations were fitted to polyexponential equations representing first-order compartmental models with first-order absorption by using nonlinear, weighted least-squares regression. All individuals were assumed to be at steady state. Some investigators have suggested that pyrimethamine pharmacokinetics are best described by a two-compartment model (17, 25); here, some subjects showed evidence of an apparent distributive phase while others did not. Furthermore, a number of subject profiles contained apparent outliers (e.g., the 16-h time point in Fig. 1d). The concentrations of outliers were verified by repeat HPLC analysis and, in some instances, by analysis using a methodologically independent enzyme inhibition assay (20) to rule out interfering substances. Other investigators have also noted substantial intraindividual fluctuations in pyrimethamine concentrations (10). Consequently, while fits of a one-compartment model were obtained for all patients, because of outlying observations and/or the lack of a clear distributive phase, it was possible to obtain a convergent nonlinear regression fit of the two-compartment model for only one patient. Thus, formal model selection techniques could not be used to identify an overall best-fitting model. Visual inspection of residual plots for the one-compartment model showed evidence of lack of fit for only three patients (see Fig. 1b, c, and d). On the basis of these considerations, a one-compartment model was used to represent the kinetics of pyrimethamine. Although for the data from 3 patients this model may have been an inadequate characterization, overall, it provided a reasonable fit to the data for all 11 subjects. The model was parameterized explicitly in terms of CL/F, V/F, and the absorption rate constant (k_a). AUC_{0-24} and $AUC_{0-\infty}$ were calculated by integrating the fitted model from 0 to 24 h and to infinity, respectively. The $t_{1/2}$ was calculated as $k_{el} = (CL/F)/(V/F)$ by using the estimated parameters for each subject.

By using a one-compartment model, a two-stage population pharmacokinetic analysis was performed (1, 6, 23); this method of population analysis is best suited to rich within-subject data. At the first stage, the model was fitted to individual-subject data by nonlinear least squares. Because serum concentrations typically exhibit heterogeneous within-subject variation, fitting usually incorporates a weighting scheme (1). Commonly, intrasubject variability is assumed to follow a proportional error (constant coefficient of variation [CV]) model, i.e., variance of a concentration at time t is modeled as proportional to $Cm(t)^2$, where $Cm(t)$ is mean concentration at t. Because of the substantial number of outlying observations at high concentrations, weighted residual plots from fits using the proportional error model still showed evidence of variance heterogeneity, suggesting that the model did not fully take this phenomenon into account. Consequently, a modified error model in which variance at t is taken to be proportional to $Cm(t)^b$ for power $b > 2$ was considered; this model allows for more profound increases in variance with mean. The power b was estimated from the data as 2.5 by standard statistical methods for the estimation of error models based on residuals from all subjects (1, 5, 6). Nonlinear weighted least-squares regression using this error model was then undertaken, and residual plots from this weighing indicated that the heterogeneity of variance was fully accommodated. At the second stage, these weighted estimates and their estimated standard errors were treated as data to estimate population characteristics (mean and standard deviation [SD]) of the pharmacokinetic model by methods previously described in detail (1, 6, 23). In this approach, a population model relating the estimate for subject i (P_i) to the population mean (P) is fitted by weighted-regression tech-

niques; i.e., the model is $P_i = P + e_i$, where e_i is a subject error incorporating components of both uncertainty in P_i due to estimation (standard errors) and variability in the population. This latter component and P are estimated from the data. Population pharmacokinetic analysis assumes that subjects arise from a population; however, the first-stage individual estimates are obtained separately for each subject, so they do not take advantage of information on the population in all the data. Thus, it is customary to obtain refined estimates of individual pharmacokinetic parameters (6, 23), which represent each subject's kinetics by a weighted combination of the first-stage estimate and the estimate of the population mean kinetics. These individual estimates are often referred to as empirical Bayes estimates and are more precise than the first-stage estimates alone, as they balance individual information against that about the population, and are preferred to first-stage estimates for characterizing individual kinetics (22). Because of this weighing, empirical Bayes estimates have the added benefit of countering the influence of individual patient outliers not enjoyed by first-stage estimates; thus, they are a natural choice for these data.

RESULTS

Course of study treatment and adverse events. Twenty-six patients meeting the eligibility criteria were enrolled in this study between November 1990 and January 1992. Sixteen subjects completed the full course of treatment; 10 discontinued treatment because of adverse experiences. Four did so because of nausea, four did so because of generalized skin eruptions (associated with fever in three, anemia in one, and headaches in one), and two did so because of headaches. An additional nine patients developed adverse experiences but completed the 21-day course of treatment. Thus, 19 of the 26 patients entering this study experienced at least one adverse event. The most common adverse events were nausea (11 patients), headaches (8), elevation of liver enzymes (7), skin eruptions (5), leukopenia (5), fever (3), sore throat (3), and elevation of creatine kinase (3 patients). Some patients experienced more than one adverse event. Four of the seven patients with increased levels of liver enzymes had mild elevations at study entry. Less frequent adverse events included anemia, hyperuricemia, hypercalcemia, hypocalcemia, hyperkalemia, light-headedness, night sweats, cough, chest pain, myalgias, earache, and blurred vision.

Serum creatinine levels increased in 23 patients after the initiation of pyrimethamine therapy. The average serum creatinine level rose from 1.08 ± 0.17 mg/dl ($n = 26$) at baseline to 1.35 ± 0.28 mg/dl ($n = 25$) after 1 week of therapy. It remained stable at 1.36 ± 0.26 mg/dl ($n = 20$) after 2 weeks and fell slightly to 1.25 ± 0.32 mg/dl after 3 weeks. Because of the long $t_{1/2}$ of pyrimethamine, substantial drug concentrations remained at the end of this study and follow-up creatinine concentrations were not obtained after complete CL of pyrimethamine.

Mean trough pyrimethamine concentrations increased from 1.58 ± 0.50 μg/ml (range, 0.45 to 2.34 μg/ml) after 1 week of therapy to 1.78 ± 0.59 μg/ml (range, 0.94 to 2.85 μg/ml) and 1.79 ± 0.57 μg/ml (range, 0.80 to 2.92 μg/ml) after 2 and 3 weeks, respectively. The difference between the mean trough values after 1 and 2 weeks was highly significant ($P < 0.0001$).

Pyrimethamine pharmacokinetics. (i) Noncompartmental analysis. Complete pharmacokinetic data for pyrimethamine were available for 12 patients. However, the profile for subject 11 was quite unusual; a sequence error in the labeling of samples and unrecorded concomitant administration of other medications are possible, but unverifiable, explanations. Although pharmacokinetic parameters were calculated for this subject, they were not comparable in magnitude to those for the remaining subjects as a consequence of the unusual pattern of observations. Accordingly, estimated pharmacokinetic parameters for this subject are excluded from Table 1, which shows the results of noncompartmental analysis for the remaining 11 subjects. The estimated pyrimethamine CLs were strongly and negatively correlated with the trough concentrations measured at time zero ($r = -0.93$) (data not shown).

TABLE 1. Pyrimethamine pharmacokinetic parameters for 11 subjects estimated by noncompartmental analysis

Subject	AUC_{0-24} (μg · h/ml)	$AUC_{0-\infty}$ (μg · h/ml)	$t_{1/2}$ (h)	CL/*F* (liters/h)	*V*/*F* (liters)	C_{max} (μg/ml)[a]
1	32.51	323.35	154.01	1.5382	341.7629	1.611
2	54.02	938.60	305.59	0.9256	408.0889	2.474
3	41.60	342.00	123.73	1.2019	214.5393	2.277
4	23.40	119.27	81.08	2.1368	249.9546	1.110
5	45.41	658.32	231.84	1.1011	368.2874	2.295
6	50.22	425.99	137.66	0.9956	197.7332	2.587
7	63.25	574.53	136.33	0.7906	155.4911	3.021
8	34.24	258.61	113.54	1.4601	239.1778	1.745
9	44.27	413.08	141.69	1.1245	229.8699	2.140
10	25.17	268.50	186.33	1.9865	534.0174	1.237
12	45.68	1,209.94	484.32	1.0945	764.7669	2.160
Mean	41.82	502.93	190.56	1.3050	336.3990	2.059
SD	12.14	324.49	115.37	0.4322	179.9485	0.581
CV	0.29	0.65	0.61	0.3311	0.5344	0.282

[a] C_{max}, maximum concentration observed.

(ii) Compartmental analysis. Fits of the one-compartment model, assuming steady state, were conducted as described above. Figure 1 shows both the first-stage weighted nonlinear regression fits and the refined, empirical Bayes fits for four patients, including the three with the most evidence of a distributive phase. The initial and refined profiles appear to be quite similar, and the estimates of CL/*F* and AUC_{0-24} were virtually identical. However, the initial and refined estimates of *V*/*F* and k_a were less consistent within individuals, particularly those with large outliers; the latter estimates downweight the influence of outlying data and are likely to be more reliable. Accordingly, we report refined estimates to characterize individual pharmacokinetics in Table 2. Because a population pharmacokinetic analysis was conducted, the means, SDs, and CVs reported in Table 2 are stage 2 estimates rather than summary statistics for these 11 subjects.

Correlation of drug exposure with adverse effects. There was insufficient evidence to suggest an association between higher drug concentrations and the incidence of adverse effects. Because of the small number of subjects in this study, it was not possible to conduct a detailed evaluation of the relationship between particular adverse effects and pyrimethamine concentrations. However, by classifying subjects as tolerant if they experienced no toxicity and intolerant if they experienced one or more adverse effects, of the 18 patients with week 1 trough levels, the 5 tolerant patients had a median trough level of 1.74 μg/ml compared with 1.68 μg/ml for the 13 subjects intolerant by week 1. At 2 weeks, the median trough levels for the 5 tolerant patients and the 11 remaining intolerant patients were 1.89 and 1.74 μg/ml, respectively; at 3 weeks, the median trough levels were 1.81 μg/ml (5 tolerant patients) and 1.87 μg/ml (6 intolerant patients). Note that five intolerant patients left this study between weeks 2 and 3. Thus, these results should be interpreted with caution, as median levels for the intolerant patients are based only on patients that had not yet left this study. Among the 11 patients remaining for pyrimethamine profiles to be obtained at week 3, the median observed maximum concentrations of pyrimethamine in serum were 2.14 and 2.23 μg/ml and the median AUC_{0-24} values were 41.39 and 42.17 μg · h/ml for the 5 tolerant and 6 intolerant patients, respectively.

Effects of intravenous drug use on pyrimethamine pharmacokinetics. To assess whether there were differences in drug

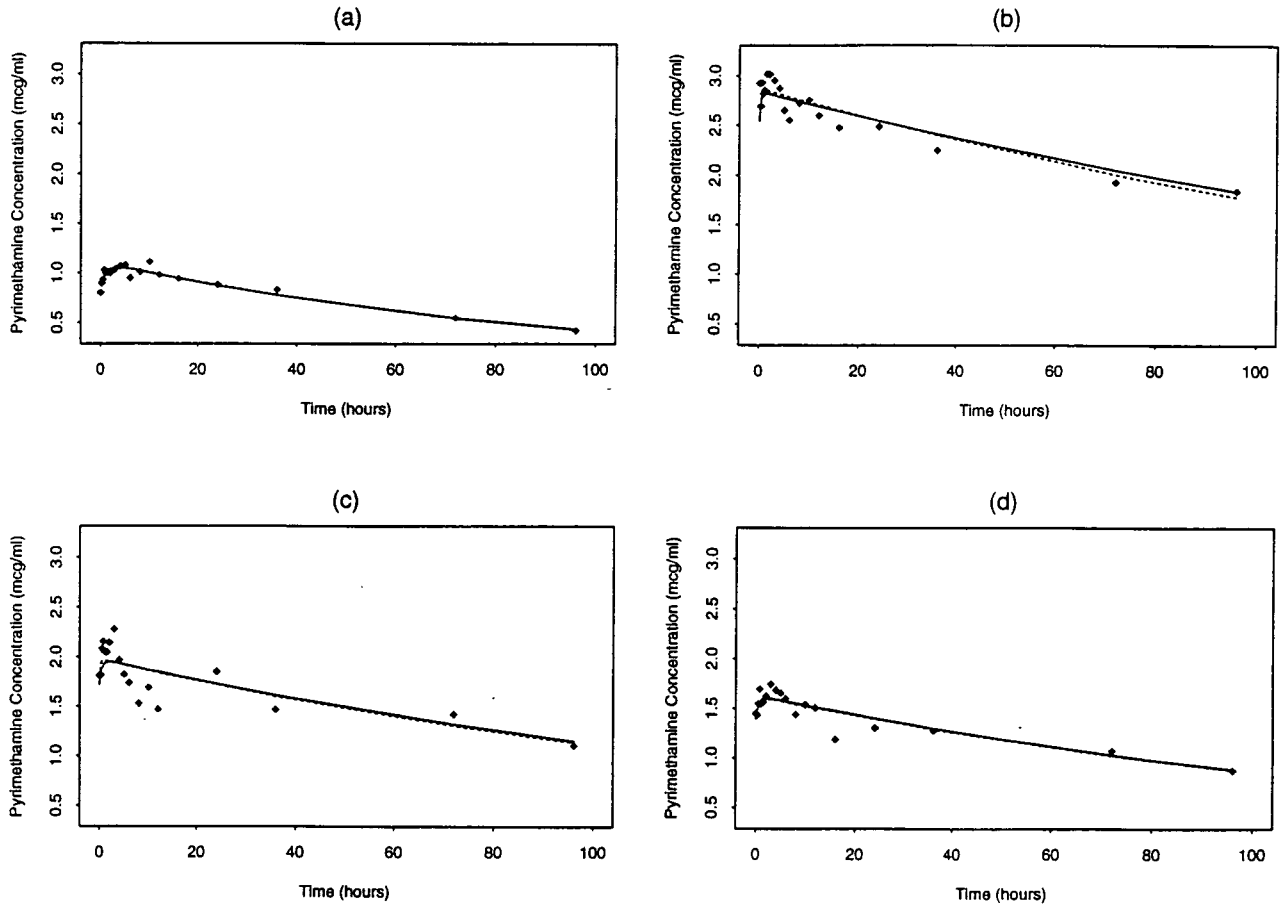

FIG. 1. Pyrimethamine pharmacokinetics at steady state. Pyrimethamine was given to subjects at 50 mg daily for 3 weeks. Plots of serum pyrimethamine concentrations at various times after administration of the final dose are shown for four representative subjects: subject 3 (a), subject 4 (b), subject 7 (c), and subject 8 (d). The calculated curve for first-stage analysis is shown as a solid line, and the refined, empirical Bayes fit is shown as a broken line.

disposition for intravenous drug users compared with nonusers, two-sample hypothesis tests (*t* test and two-sided Wilcoxon test) were applied to the mean differences of each pharmacokinetic parameter obtained from both noncompartmental and compartmental analyses. For neither analysis was the evidence strong enough to suggest a difference between the two groups in any mean pharmacokinetic parameter value (data not shown).

TABLE 2. Pyrimethamine pharmacokinetic parameters for 11 subjects estimated by compartmental analysis

Subject	AUC_{0-24} (μg · h/ml)	$AUC_{0-\infty}$ (μg · h/ml)	$t_{1/2}$ (h)	CL/*F* (liters/h)	*V*/*F* (liters)	k_a (liters/h)
1	33.97	289.25	132.30	1.4716	280.8810	0.7872
2	52.83	611.80	183.75	0.9464	250.8733	1.4976
3	44.21	352.20	123.67	1.1310	201.7729	1.8278
4	23.34	113.14	71.25	2.1426	220.2571	0.8166
5	47.76	539.17	178.79	1.0469	270.0237	1.1929
6	50.30	435.12	135.12	0.9940	193.7630	2.2267
7	64.53	622.29	151.76	0.7748	169.6262	3.6241
8	36.19	255.72	108.52	1.3816	216.3202	1.3231
9	46.07	413.15	139.93	1.0852	219.0596	1.6533
10	24.49	199.10	124.88	2.0418	367.8754	0.3522
12	45.57	524.92	182.50	1.0971	288.8637	1.0088
Mean	42.66	395.90	139.32	1.2798	245.6278	1.5478
SD	12.29	169.84	34.14	0.4141	63.8332	1.3394
CV	0.29	0.43	0.25	0.3235	0.2598	0.8653

Analysis of zidovudine-pyrimethamine-pharmacokinetic interaction. Zidovudine concentration-time profiles before and after the initiation of pyrimethamine treatment were available for 11 patients. The profile of one patient was highly irregular and was not included in this analysis. The population values for zidovudine pharmacokinetic parameters without and with pyrimethamine treatment are given in Table 3. Pharmacokinetic profiles were obtained for all but one of the 10 patients listed in Table 2 after a 100-mg dose both before and after pyrimethamine; the remaining subject received a 200-mg dose on both occasions, and concentrations were corrected to a dose of 100 mg for this patient. These values fall within the ranges of values reported for zidovudine by other investigators. When values without and with pyrimethamine were compared, no significant differences in any of these parameters were detected by the paired *t* test and the signed rank test (data not shown).

DISCUSSION

The primary focus of this study was to evaluate the pharmacokinetics of pyrimethamine administered daily to HIV-infected patients on concurrent zidovudine therapy. A particular concern was to confirm previous reports suggesting an increased CL and a shorter $t_{1/2}$ for pyrimethamine in HIV-infected patients compared with those in uninfected persons, since this could have a major impact on dosing recommendations. Another objective was to determine whether the high

TABLE 3. Summary statistics for zidovudine pharmacokinetic parameters determined by noncompartmental analysis before and after coadministration of pyrimethamine[a]

Treatment	AUC_{0-4} (μg · h/ml)	$AUC_{0-\infty}$ (μg · h/ml)	k_{el} (liters/h)	$t_{1/2}$ (h)	CL/*F* (liters/kg/h)	*V*/*F* (liters/kg)	C_{max} (ng/ml)[b]
Without pyrimethamine							
Mean	869.11	1,265.42	0.6163	1.4132	1.9820	3.4803	651.55
Median	773.25	878.61	0.6110	1.1346	1.9196	3.0390	580.68
SD	440.88	1,360.86	0.2088	1.0850	0.8159	1.4058	361.60
CV (%)	50.7	107.5	33.9	76.8	41.2	40.4	55.5
With pyrimethamine							
Mean	992.27	1,224.54	0.5583	1.4617	1.7654	3.9472	799.43
Median	821.74	1,069.82	0.6576	1.0553	1.7879	2.5903	587.41
SD	530.44	713.77	0.2031	0.6858	0.7722	3.3578	606.32
CV (%)	53.5	58.3	36.4	46.9	43.7	85.1	75.84

[a] Based on data for 10 subjects after a 100-mg oral dose of zidovudine.
[b] C_{max}, maximum concentration observed.

rate of adverse effects in patients with AIDS could be related to unusual pharmacokinetics.

After 3 weeks of treatment with pyrimethamine at 50 mg/day, steady-state trough levels ranged from 0.8 to 2.92 μg/ml. This range compares with steady-state trough levels of 1.3 to 4.4 μg/ml seen within patients not infected with HIV who were treated with 25 mg of pyrimethamine twice a day (10). The latter study used a bioassay and may have measured active metabolites, thus overestimating pyrimethamine concentrations. The finding that the trough levels in HIV-infected and uninfected patients on similar regimens are similar is consistent with the absence of significant differences in the pharmacokinetic parameters of HIV-infected patients.

Despite the use of a loading dose of 200 mg of pyrimethamine, the mean trough level rose significantly between 7 and 14 days of therapy, from 1.58 to 1.77 μg/ml, and then stabilized. In retrospect, this delay in achieving steady-state concentrations is not surprising. On the basis of the average $t_{1/2}$ of 139 h, achieving steady state should require about 4 weeks in the absence of a loading dose. At that time, peak concentrations are expected to be about ninefold higher than the peak levels after a single 50-mg dose. On the basis of the average *V* of 245 liters, a loading dose of 500 mg would be required to achieve an initial peak level of approximately 2 μg/ml. Because such a loading dose might prove intolerable, an alternate approach might be to front-load with an accelerated initial regimen of 200 mg/day for the first 3 days. These approaches have not been tried in patients, and it is not clear whether they could lead to an increased incidence of adverse effects.

Noncompartmental and compartmental pharmacokinetic analyses were performed. The results for the CL and AUC_{0-24} (a measure of the drug exposure attributable to a single dose at steady state) were remarkably consistent for these two analyses, both for the population and for individual subjects. Both analyses provided an estimated mean CL of 1.3 ± 0.4 liters/h and an AUC_{0-24} of 42 ± 12 μg · h/ml. For other pharmacokinetic parameters, these two analyses provided disparate estimates. This may be due in part to the paucity of information on k_{el}, accurate estimation of which requires observations extending over several half-lives. Final observations were taken at 96 h on the basis of previously reported pyrimethamine half-lives in HIV-infected patients (11, 24); however, both analyses yielded an estimated mean $t_{1/2}$ exceeding 96 h.

The fact that the previously reported short half-lives were based on even shorter observation intervals of 10 to 48 h (11, 24) is of concern. If pyrimethamine has a significant distribution phase, so that pharmacokinetics are better described by a two-compartment model, apparent half-lives measured over short intervals would be likely to reflect a composite of distribution and $t_{1/2}$. Consistent with this, half-lives computed from peak and trough levels were much shorter for a dosage of 50 mg/day than for a dosage of 100 mg/day. Although a distributive phase was not evident in many patients in the current study, several patients did exhibit an apparent distributive phase (e.g., Fig. 1b, c, and d), which would not be adequately represented by a one-compartment model. However, our use of a forecast interval approach to determine those points to be included in the calculation of the terminal elimination rate constant should have resulted in the rejection of points significantly affected by a distribution phase component.

From the population pharmacokinetic analysis based on the one-compartment model, the mean pyrimethamine $t_{1/2}$ was estimated as 139 ± 34 h, the AUC_{0-24} was 42.7 ± 12.3 μg · h/ml, the CL was 1.28 ± 0.41 liters/h, the *V* was 246 ± 64 liters, and the k_a was 1.5 ± 1.3 liters/h. These parameter values are similar to those of uninfected individuals, but with a somewhat longer $t_{1/2}$ resulting principally from a lower CL. With the exception of the k_a (which is usually difficult to estimate precisely in the fitting of nonlinear compartment models under standard sampling protocols), the pharmacokinetic variability of pyrimethamine is of the order of magnitude typical of most drugs.

From the limited data available, no exceptionally large differences between the intravenous-drug-using and non-drug-using HIV-infected populations emerged. There was no evidence of a major pharmacokinetic interaction between pyrimethamine and zidovudine. Although these conclusions are limited by the small number of patients for whom data were available, very large differences would have been detected, even if formal significance was not achieved.

An unexpected finding of our study was a substantial rate (73%) of adverse experience in the 3-week regimen, which employed the standard 50-mg daily dose that is used for at least 6 weeks in the treatment of toxoplasma encephalitis. Ten of 26 patients (38%) permanently discontinued the study medication because of nausea, headaches, or skin rash. An additional nine patients (35%) experienced one of these adverse effects. There were no differences between intravenous drug users and nonusers in the likelihood of having an adverse experience.

The observed range of pyrimethamine concentrations was not substantially different from that seen in non-HIV-infected persons. Because there was nothing to suggest that the study

patients differed from the general HIV-infected population, our results imply that this population may be relatively intolerant of pyrimethamine in the doses usually used to treat toxoplasma encephalitis. Up to 40% of HIV-infected patients with toxoplasma encephalitis treated with either pyrimethamine-sulfadiazine or pyrimethamine-clindamycin have had adverse drug reactions that required a change of therapy (3, 8). Previously, skin rashes were usually thought to be related to the clindamycin or sulfonamide component. In part, this was supported by the observation that the rash usually resolved when the patient was switched to the alternative treatment regimen also containing pyrimethamine. However, the rash may resolve despite continuation of the original regimen. This occurred in one of our patients. Ruf et al. studied the pyrimethamine-sulfadoxine combination to prevent relapses of toxoplasma encephalitis in 56 HIV-infected patients (21). Of 23 patients who developed a rash or pruritus, 19 elected to continue therapy with the same drugs. In all 19 patients, the skin manifestations resolved.

Much of the previously reported intolerance to pyrimethamine-sulfonamide or pyrimethamine-clindamycin in patients with AIDS may in fact be related to pyrimethamine. Switching to the alternate regimen may not always be necessary. Further studies are needed to determine whether treating through these adverse events, particularly skin rash, is feasible.

The frequencies of disabling nausea or vomiting and headaches were higher than those reported previously. These symptoms were not associated with higher serum pyrimethamine concentrations or with the presence of abnormal liver chemistries at baseline. Hematologic toxicity was uncommon during this 3-week study. The coadministration of folinic acid may have helped protect the bone marrow of patients.

We observed that pyrimethamine caused a predictable increase in serum creatinine levels, confirming the findings of Opravil et al. (19), who demonstrated that this increase resulted from the inhibition of renal tubular secretion of creatinine by pyrimethamine. Clinicians should be aware that a mild elevation in the serum creatinine level of a patient receiving pyrimethamine usually does not indicate renal damage.

In conclusion, in this first formal pharmacokinetic study of pyrimethamine in HIV-infected patients, there was great variability in the concentrations of pyrimethamine in serum achieved with a dosage of 50 mg daily, reflecting the variability in CL rates. This suggests that drug level monitoring might be useful during pyrimethamine therapy of toxoplasma encephalitis. However, additional studies are needed to determine whether a therapeutic range can be established. In addition, the steady-state drug levels were comparable to those in similarly treated patients without HIV infection and the pyrimethamine CL was no more rapid than in uninfected individuals. No changes in the standard pyrimethamine treatment regimens appear to be necessary for HIV-infected patients. The $t_{1/2}$ was typically more than 3 days. This suggests that a large loading dose or an accelerated initial dosing regimen may be desirable to rapidly achieve steady-state levels and that subsequent dosing need not be daily. A need for further investigation of such dosing regimens appears to be indicated.

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