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Intravenous and Oral Zidovudine Pharmacokinetics and Coagulation Effects in Asymptomatic Human Immunodeficiency Virus-Infected Hemophilia Patients

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Pharmacokinetic and coagulation studies were carried out over a 12-week period with 11 asymptomatic hemophilia patients with human immunodeficiency virus infection receiving zidovudine (ZDV). The patients received 300 mg every 4 h while awake (the accepted dose at the time of this study); consecutive 24-h intravenous (i.v.) and 12-h oral pharmacokinetic studies were conducted at weeks 1, 6, and 12. Coagulation studies were conducted at weeks 0, 4, 8, and 12. The numbers of units of factors VIII and IX and cryoprecipitate transfused during the 12-week periods before, during, and after ZDV treatment were recorded. Following i.v. and oral ZDV administration, the concentration in plasma declined rapidly over the first 4 h, and in some patients, ZDV was still detectable at 4 to 10 h. The i.v. total clearances (means ± standard deviations) were 14.9 ± 7.3, 11.2 ± 3.7, and 15.1 ± 4.7 ml/min/kg of body weight. The i.v. distribution volumes were 1.08 ± 0.5, 1.0 ± 0.4, and 1.65 ± 1.4 liters/kg. The bioavailabilities were 0.54 ± 0.22, 0.46 ± 0.19, and 0.59 ± 0.13 at weeks 1, 6, and 12, respectively. The pattern of ZDV-glucuronide (GZDV) disposition was similar to that of ZDV, and the peak plasma GZDV-to-ZDV ratio was higher after oral dosing, consistent with first-pass metabolism. In some individuals, up to 33% of an i.v. dose was excreted unchanged. At weeks 6 and 12, >300 mg of total ZDV (GZDV plus ZDV) was recovered in the urine of some patients, suggesting tissue redistribution. Concentrations in plasma after oral ZDV administration were variable, both within and between patients. The von Willebrand antigen level consistently decreased throughout the study but was not accompanied by a parallel change in ristocetin cofactor A activity, and no clinical adverse effects on coagulation were noted. This study demonstrates that ZDV can be used in hemophilia patients without worsening of their bleeding tendencies. The clinical significance of decreased ZDV clearance and the prolonged terminal elimination phase of ZDV will require further study with patients receiving chronic ZDV.

Many hemophilia patients have previously contracted human immunodeficiency virus (HIV) infection from contaminated factor VIII products (7, 16, 27). Continued monitoring of asymptomatic, HIV-infected hemophilia patients indicates that clinical manifestations of HIV disease will probably develop, with ultimate progression to AIDS-related complex (ARC) or AIDS, in a large percentage of these patients (8, 9, 13). Therefore, the chronic use of zidovudine (ZDV) or other antiretroviral agents is a likely scenario for many hemophilia patients (14).

ZDV treatment is currently being evaluated as monotherapy and in combination with didanosine or zalcitabine treatment of hemophilia patients with HIV infection to assess the impact of treatment on the progression of disease. Prior to initiation of these multicenter studies, a preliminary phase I evaluation of ZDV pharmacokinetics and coagulation effects in hemophilia patients was conducted under the aegis of the AIDS Clinical Trials Program of the National Institute for Allergy and Infectious Diseases AIDS Program. We have previously reported single-dose (20) and multiple-dose (21) oral pharmacokinetic data from this trial. The present report extends the preliminary data on oral ZDV treatment to provide (i) a comparison of the pharmacokinetics of ZDV following intravenous (i.v.) and oral administration on three separate occasions over a 12-week dosing period, (ii) an evaluation of the disposition of ZDV-glucuronide (GZDV) during multiple dosing, (iii) an assessment of the patterns of renal excretion of ZDV and GZDV during multiple dosing, and (iv) an evaluation of the influence of prolonged ZDV administration on coagulation parameters in asymptomatic, HIV-infected hemophilia patients.

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MATERIALS AND METHODS

Hemophilia patients (and two nonhemophiliac controls for coagulation studies) with HIV infection were admitted to the study after written informed consent was obtained. HIV infection was determined by two consecutively positive enzyme-linked immunosorbent assays and a positive confirmatory Western blot (immunoblot) test. No patient had severe cardiopulmonary, gastrointestinal, or renal disease, and none of the hemophiliacs were symptomatic from their HIV infection. In addition, the patients were not receiving chronic medications suspected of interfering with ZDV metabolism. Baseline hematologic, chemistry, and clotting studies were performed prior to drug administration and then

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every 4 weeks thereafter. Entry criteria included the following: hematocrit, >30; hemoglobin, ≥9 g%; granulocyte count, ≥1,000 cells per mm^3; platelet count, ≥50,000/mm^3; aspartate transaminase (AST) level, ≤5× upper limit of normal; and serum creatinine, ≤1.5× upper limit of normal.

Pharmacokinetic studies. Patients were admitted to the Clinical Research Center at the University of Rochester on three separate occasions (day 1, week 6, and week 12) for a 36- to 48-h period. At the initiation of each study period, the bladder was emptied and an i.v. catheter was placed in each arm. The patency of the venous access sites was maintained with a dilute heparin solution. ZDV (300 mg) was administered i.v. in 100 ml of 5% dextrose in water over 1 h. Blood samples were collected at 0, 0.5, 1, 2, 3, 4, 6, 8, 10, 12, and 24 h. Urine was collected every 4 h for 12 h and then again at 24 h. Each patient then received a single 300-mg oral dose (three 100-mg capsules), with blood and urine collection repeated as described above except that the collection period was 12 h. All patients took nothing orally for 2 h prior to the dose and for 1 h after the dose. It should be noted that at the time this study was conducted, 1,200 mg/day was the recommended dose. The patients were discharged and instructed to take 300 mg of ZDV every 4 h (1,200 mg/day) while awake, 1 h before a meal if possible. After 6 and 12 weeks, i.v. and oral pharmacokinetic studies were repeated as described above. During some i.v. studies, the blood sample collection protocol was inadvertently begun at the end of the 1-h i.v. infusion period. For these patients, estimation of the ZDV concentration at the end of the infusion was calculated by linear regression utilizing the mean slope of the initial linear α-phase decline determined from the i.v. study periods in which the sampling was begun at the beginning of the infusion. For all studies, plasma (obtained by centrifugation) and urine were stored at −70°C until assayed.

ZDV and GZDV were measured by the previously reported high-performance liquid chromatography (HPLC) method of Good et al. (10). Plasma samples were incubated for 3 h at 56°C to inactivate the virus. Stability studies of ZDV and GZDV were conducted which indicated that no degradation occurred during the inactivation period. To 0.5 ml of patient sample or serum standard (0.1 to 20 μM [39- to 5,000-ng/ml] analytical grade ZDV–0.2 to 40 μM [78- to 10,000-ng/ml] GZDV), 0.1 ml of internal standard (BW-A22U; Burroughs Wellcome Laboratories, Research Triangle Park, N.C.) was added. The sample was then allowed to stand for 15 min to attain equilibrium. ZDV and GZDV were extracted from plasma with a liquid-solid extraction phase technique. The 3-ml extraction columns (C-18; Analytichem International, Harbor City, Calif.) were conditioned with one reservoir volume of methanol followed by 2 volumes of phosphate-buffered saline (PBS). The vacuum was adjusted to 4 to 7 in. (10.16 to 17.78 cm) of Hg and controlled with a plastic stopcock. Plasma samples were then transferred to the individual extraction columns and washed with 1 ml of PBS. The columns were then dried for 5 min under full vacuum. ZDV and GZDV were eluted with two 1-ml rinses with methanol and evaporated to dryness under nitrogen in a water bath at 37°C. Samples were reconstituted with 200 μl of 15% acetonitrile in water and injected (50 to 150 μl) onto the HPLC system.

The HPLC system consisted of a Varian 5000 pump (Varian Laboratories, Walnut Creek, Calif.), a Waters variable-wavelength detector, and a 710B automatic injector (Waters Associates, Milford, Mass.). The composition of the mobile phase was adjusted with a gradient elution program, which consisted of ammonium phosphate buffer-acetonitrile (90:10, vol/vol) for 26 min, a linear change to 30% acetonitrile for 0.3 min, 30% acetonitrile for 9 min, and then a linear change back to ammonium phosphate buffer-acetonitrile (90:10) over 3 min, which was then continued for 15 min. The mobile phase was run at 1.3 ml/min, and ZDV and GZDV were detected at 267 nm. Urine samples were diluted 1:5 or 1:10 with PBS. A 0.5-ml aliquot was filtered through a 0.22-μm-pore-size filter (Millipore, Milford, Mass.), and 5 to 15 μl was injected onto the HPLC column. ZDV and GZDV were eluted with ammonium phosphate buffer-acetonitrile (90:10) in an isocratic manner at a flow rate of 1.3 ml/min. ZDV and GZDV concentrations were determined by calculation of the ratio of the corresponding peak area to the area of the internal standard. Linear regression was used to determine the unknown sample concentrations. High-concentration (2,000- and 4,000-ng/ml) and low-concentration (200- and 800-ng/ml) quality control samples for ZDV and GZDV were analyzed with each set of patient samples and yielded an interday coefficient of variation of <10% for all quality control specimens. The lower limits of detection for ZDV and GZDV were 20 and 40 ng/ml, respectively.

The area under the concentration-time curve (AUC) for plasma and the area under the first moment of the concentration-time curve (AUMC) were determined by polynomial interpolation with the LAGRAN computer program (26). The mean residence time (MRT) was calculated as AUMC/AUC − 1/2T, where T is infusion time. After i.v. dosing, clearance (CL) was calculated as dose/AUC and the volume of distribution at steady state (V_{ss}) was calculated as CL × MRT. The plasma drug concentration data were graphed on a log-linear plot, and the terminal elimination phase was determined by visual inspection of the curve. The apparent oral-dose clearance (CL_{oral}) was calculated as dose/AUC, where dose was equal to 300 mg. The elimination phase half-life ($t_{1/2}$) was calculated as $0.693/\lambda_z$, where λ_z is the slope of the terminal elimination phase. Renal clearance (CL_R) was calculated as the amount of drug recovered in the urine from time zero to time t divided by the corresponding AUC. In order to more accurately characterize the prolonged elimination pattern that was noted, the data from patients with measurable concentrations in plasma at up to 9 h were fit to a two-compartment, bolus model with nonlinear least-squares regression analysis (PCNONLIN). Because the data did not describe the infusion time sufficiently, an attempt was made to obtain an accurate fit of the time points from 1 to 10 h after the dose. Only patients that had received an i.v. dose had high enough concentrations in plasma to allow for compartmental analysis. While the oral data revealed a late phase of elimination, the datum points were insufficient to characterize this finding with nonlinear regression.

Coagulation studies. The following laboratory parameters of coagulation were measured for each patient before (at week 0) and during (at weeks 4, 8, and 12) ZDV treatment: factor VII, factor VIII, factor IX, prothrombin time (PT), partial thromboplastin time (PTT), ristocetin cofactor A, von Willebrand antigen, fibrinogen, euglobulin clot lysis time, and factor VIII inhibition assay. Platelet count and bleeding time were measured approximately weekly. Routine techniques were used to measure the coagulation parameters (30). The hemophilia center provided the coagulation factor use data during the 12-week periods prior to, during, and after ZDV treatment.

Depending on the parameter, coagulation test results were not used in the analysis if a factor or cryoprecipitate transfusion was given within 1 to 2 days prior, because of the possible effect on the results. For example, factor VIII levels

TABLE 1. Demographic data for 11 asymptomatic, male HIV-infected hemophilia patients upon entry into the study

Patient	Age (yr)	Wt (kg)	Serum creatinine (mg/dl)	AST U/liter	T4 level (no./mm^3)
1	43	94	1.0	59	145
2	27	65	0.8	50	221
3	46	60	0.7	41	176
4	25	100	0.8	50	502
5	31	57	0.6	86	103
6	34	63	0.6	220	188
7	35	63	0.8	71	103
8	33	88	0.9	104	189
9	36	88	0.7	292	204
10	29	80	0.8	36	171
11	23	67	0.8	180	3
Mean ± SD	33 ± 7	75 ± 15	0.8 ± 0.1	108 ± 85	182 ± 123

would be expected to be much higher after factor VIII transfusion.

To evaluate the effect of ZDV, for each coagulation parameter, we compared the last test value obtained while the patient was taking ZDV (usually week 12) to that at week 0 (pre-ZDV). The null hypothesis—that there was no change between, prior to, and during ZDV treatment—was tested by the Wilcoxon signed-rank test for matched variables. Spearman's coefficient of correlation was used to describe relationships between quantitative variables. A two-tailed P value equal to or less than 0.05 was considered significant. P values are reported without adjustments for multiple comparisons.

RESULTS

Patients. Eleven asymptomatic hemophilia patients (all CDC class II, except 1 patient who was class IV E) were admitted into the study. All 11 completed the first i.v. pharmacokinetic study, but 2 patients dropped out prior to the first oral dose study. Nine patients completed the first 6 weeks of ZDV therapy and participated in both the i.v. and oral pharmacokinetic studies at week 6. One patient (patient 5) was excluded from the study at week 8 because of increasing liver enzyme levels, possibly associated with ZDV therapy. Two patients (patients 2 and 7) participated in the i.v. pharmacokinetic study at week 12 but not the oral segment as a result of noncompliance.

Coagulation studies were completed for 10 of the HIV+ asymptomatic hemophilia patients. Of the eight with hemophilia A (factor VIII deficiency), seven were severely deficient and one was moderately deficient. One patient had severe hemophilia B (factor IX deficiency), and one had severe von Willebrand disease. The von Willebrand disease patient and one hemophilia A patient received regularly scheduled cryoprecipitate or factor VIII transfusions prophylactically, and one hemophilia A patient received prophylactic transfusions perioperatively (knee surgery). The other patients received factor VIII or IX transfusions only when needed. Two HIV+ symptomatic (ARC) nonhemophiliacs served as controls. Patient 5 had the usual four sets of coagulation data collected, but the data were collected on weeks 0, 4, 6, and 8 instead of weeks 0, 4, 8, and 12.

As can be seen from Table 1, all 11 patients had elevated AST levels upon entry. Patient 5, who was later discontinued from the study, had a progressive increase in AST level over the 12-week period, but the AST level eventually returned to baseline following cessation of ZDV treatment. No other adverse events were noted during the study period. The demographic data are summarized in Table 1.

i.v. ZDV pharmacokinetics. Following i.v. administration, concentrations of ZDV and GZDV in plasma declined in a multiexponential fashion (Fig. 1). The ZDV concentrations (means ± standard deviations) 1 h after the end of i.v. infusion were 1,245 ± 536 (week 1), 1,149 ± 330 (week 6), and 737 ± 289 (week 12) ng/ml. ZDV concentrations declined rapidly and were below the assay sensitivity level in most patients after 4 to 6 h. However, in some patients, ZDV and GZDV were detectable between 6 and 10 h. In patients with measurable concentrations in plasma up to the 9-h time point, the data were sufficient to be fit to a two-compartment, bolus model (see Table 4). It should be noted that although the early time points were not able to be fit well (thus the need for a bolus model instead of an infusion), the points from 1 to 9 h gave good datum fits for some of the patients after i.v. dosing. The main determinant of a good datum fit was that the ZDV concentration be measurable up to 9 h. Thus, while a prolonged elimination phase was visually apparent after 4 h, in other i.v. profiles as well as with the oral data, the data did not fit well to this model. It was apparent that the reason the i.v. data fit the model while the oral data did not was because the entire dose was systemically available, resulting in higher concentrations in plasma for a longer period than that observed after oral dosing.

The ZDV CLs at weeks 1, 6, and 12 were similar, with values of 14.9 ± 7.3, 11.2 ± 3.7, and 15.1 ± 4.7 ml/min/kg (Table 2). However, two patients (patients 2 and 8) had a 1.5- to 2-fold increase in CL from week 1 to week 12, and patient 5 had a 50% decrease in ZDV CL associated with an increasing AST level. The V_{ss}s were 1.08 ± 0.5, 1.0 ± 0.4, and 1.65 ± 1.4 liters/kg. The overall elimination $t_{1/2}$s were 2.1 ± 1.9, 2.0 ± 1.5, and 3.2 ± 3.5 h. However, in some patients, the $t_{1/2}$ was longer because of the presence of measurable ZDV concentrations after 4 h. The renal elimination of ZDV and GZDV was variable within and between patients. The ZDV CL_Rs were 2.3 ± 1.7, 2.6 ± 1.6, and 2.1 ± 1.2 ml/min/kg, and the GZDV CL_Rs were 13.8 ± 8.7, 11.6 ± 5.5, and 17.0 ± 8.2 ml/min/kg, respectively. Most of the ZDV and GZDV was excreted in the urine during the 0- to 4-h period. However, for many patients, considerable amounts of GZDV were recovered from the later urine collections. Pharmacokinetic parameters for ZDV, determined by noncompartmental and nonlinear regression, are summarized in Tables 2 and 3, respectively. Figure 2 illustrates the patterns of urinary recovery of ZDV and GZDV after i.v. administration.

Oral pharmacokinetics. Because of the nature of the study design, the initial concentrations of ZDV and GZDV in plasma at time zero (time 24 h of the i.v. study) were below the assay sensitivity level at weeks 1, 6, and 12. Following the oral ZDV study dose (300 mg), peak ZDV and GZDV concentrations occurred at 0.5 to 1.0 h. The peak ZDV concentrations were 1,733 ± 949, 1,018 ± 330, and 1,025 ± 481 ng/ml at weeks 1, 6, and 12, respectively. The decay pattern of ZDV concentrations was rapid from 1 to 4 h. However, in five patients at week 1, all patients at week 6, and three patients at week 12, ZDV was detectable after 4 h, with a prolonged elimination pattern. As mentioned above, this pattern was noted in some patients after i.v. administration as well. The CL_{oral}s were 29 ± 14, 27 ± 9, and 26 ± 11 ml/min/kg for the three oral study days, respectively. The bioavailabilities were 0.54 ± 0.22, 0.46 ± 0.19, and 0.59 ± 0.13 for the three oral study days, respectively. The phar-

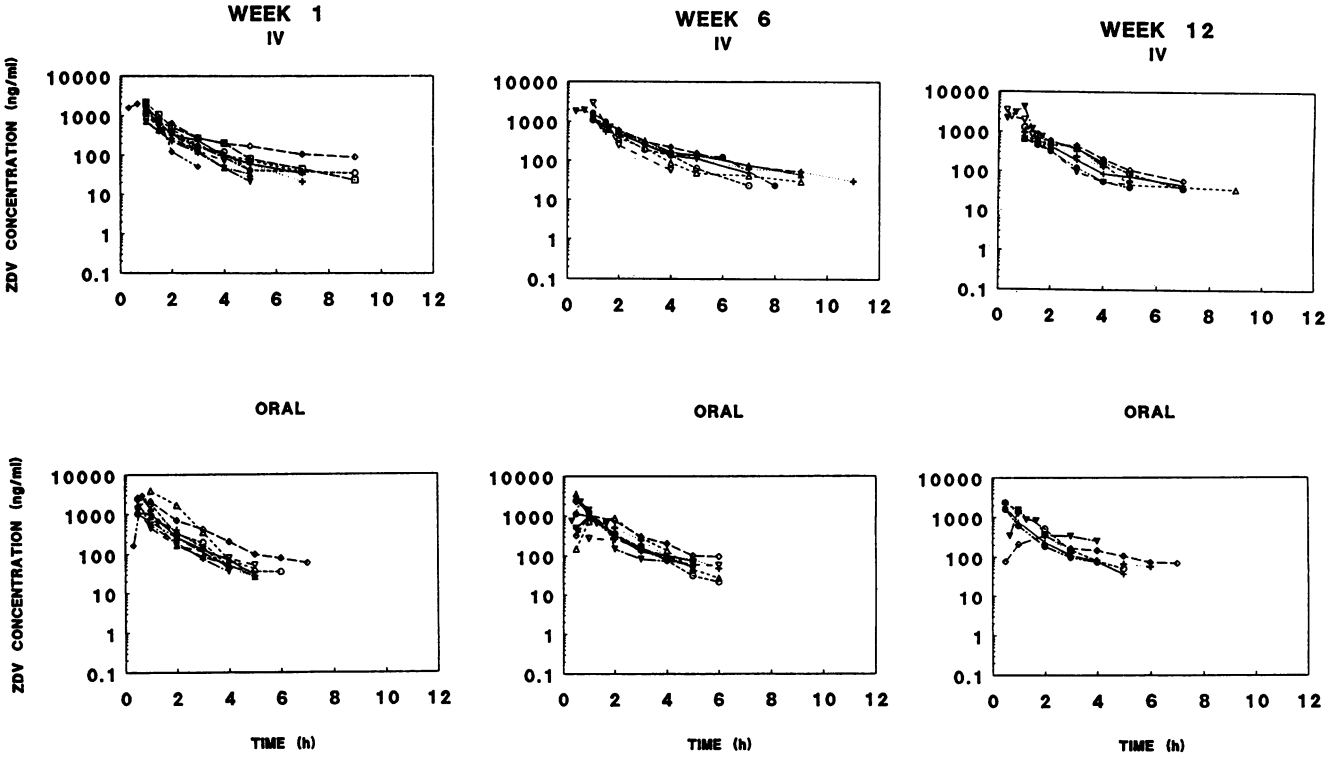

FIG. 1. Plasma concentration-versus-time profiles for ZDV among hemophilia patients (each symbol represents one patient) following i.v. and oral dosing at weeks 1, 6, and 12.

macokinetic parameters obtained during oral ZDV administration are summarized in Table 4.

The 12-h urinary recoveries of ZDV following oral administration ranged from 14 to 55 mg at week 1 to 28 to 55 mg at week 6 and 20 to 52 mg at week 12. In comparison, the total urinary recoveries of ZDV [ZDV + (GZDV × 267.2/442.4)] ranged from 206 to 333 mg at week 1 to 160 to 330 mg at week 6 and 173 to 389 mg at week 12 (Table 5).

Coagulation studies. In the 10 HIV$^+$ hemophiliacs and 2 HIV$^+$ nonhemophiliac controls, there was no evidence of an effect of ZDV on any of the following coagulation parameters: factor VII, factor VIII, factor IX, PT, PTT, von Willebrand antigen, ristocetin cofactor activity, fibrinogen level, and bleeding time (Table 6). Also, the euglobulin clot lysis time and the factor VIII inhibition assay results remained normal and constant.

The median increase in platelet count from before ZDV treatment to during ZDV treatment was 44,000/mm^3, a statistically significant change ($P = 0.007$) (Table 6). Of note, this increase was seen in all but two patients, and these two patients started with platelet counts greater than 240,000/mm^3 and had a trivial drop of less than 8,000/mm^3.

TABLE 2. Pharmacokinetic parameters for ZDV[a]

Patient	Wk 1				Wk 6				Wk 12			
	CL (ml/min/kg)	CL_R (ml/min/kg)	V_{ss} (liters/kg)	$t_{1/2}$ (h)	CL (ml/min/kg)	CL_R (ml/min/kg)	V_{ss} (liters/kg)	$t_{1/2}$ (h)	CL (ml/min/kg)	CL_R (ml/min/kg)	V_{ss} (liters/kg)	$t_{1/2}$ (h)
1	7.4	0.7	0.54	1.5	10.5	2.4	1.06	1.9	13.9	1.7	1.63	3.0
2	12.4	2.5	1.02	2.1	13.1	2.9	1.45	5.5	16.7	0.7	4.58	11.0
3	12.5	2.7	1.10	3.7	14.3	2.5	1.08	1.2	11.6	3.7	0.95	1.4
4	9.4	1.7	0.67	1.3	8.0	2.7	0.87	2.4	10.0	1.5	0.85	1.8
5	24.7	3.6	1.67	1.0	10.8	2.6	1.04	2.4	NS[b]	NS	NS	NS
6	10.6	1.3	0.63	1.0	10.0	0.7	0.73	0.9	24.0	NC[c]	1.78	1.2
7	20.1	0.8	1.41	1.1	6.7	1.1	0.29	1.0	19.3	0.7	2.40	5.0
8	6.9	1.4	1.65	7.1	8.5	2.0	0.93	2.3	12.2	2.7	0.14	1.7
9	5.6	1.4	0.40	2.2	NS	NS	NS	NS	NS	NS	NS	NS
10	18.9	3.2	1.21	0.8	18.8	6.5	1.53	0.8	13.1	3.4	0.88	0.6
11	26.3	6.4	1.54	0.8	NS	NS	NS	NS	NS	NS	NS	NS
Mean ± SD	14.9 ± 7.3	2.3 ± 1.7	1.08 ± 0.5	2.1 ± 1.9	11.2 ± 3.7	2.6 ± 1.6	1.00 ± 0.4	2.0 ± 1.5	15.1 ± 4.7	2.1 ± 1.2	1.66 ± 1.4	3.2 ± 3.4

[a] Parameters were obtained with noncompartmental analysis at weeks 1, 6, and 12 following i.v. ZDV (300 mg) administration.
[b] NS, not studied.
[c] NC, urine not collected.

TABLE 3. Pharmacokinetic parameters in patients with sufficient plasma concentration datum points to allow for nonlinear regression analysis[a]

Patient	Wk	$t_{1/2\alpha}$ (h)	$t_{1/2\beta}$ (h)	k_{12} (h^{-1})	k_{21} (h^{-1})
2	6	0.49	2.2	0.36	0.53
3	1	0.44	2.9	0.53	0.42
	6	0.64	2.0	0.18	0.50
4	6	0.80	4.8	0.24	0.24
5	6	0.59	3.0	0.33	0.41
8	1	0.78	26.7	0.58	0.10
	6	0.69	2.9	0.27	0.46
9	1	0.36	1.9	0.60	0.69
Mean ± SD		0.60 ± 0.16	5.8 ± 8.5	0.39 ± 0.16	0.42 ± 0.18

[a] Parameters were obtained following i.v. ZDV treatment, using a two-compartment model for data analysis. k_{12} and k_{21}, intercompartmental transfer rate constants.

The von Willebrand antigen level was also significantly affected by ZDV treatment, with a median drop of −80% (P = 0.008). However, the median baseline von Willebrand antigen level was high (220%) and fell to 103%, which is within normal limits in our laboratory (60 to 150%).

We also examined the weekly requirements for transfusion of factor VIII (eight patients), factor IX (one patient), and cryoprecipitate (one patient) among the hemophiliacs. Before, during, and after ZDV treatment, the median numbers of units transfused per week (ranges in parentheses) were 1,585 (O to 3,951), 388 (0 to 2,923), and 601 (0 to 2,794), respectively. Assuming that if ZDV was to have a beneficial effect there should be fewer units transfused during ZDV treatment than either before or after, for each patient, we calculated the transfusion requirement as (units transfused during treatment − units transfused before treatment) + (units transfused during treatment − units transfused after treatment). On average, these sums of changes should be null if no treatment effect was to exist, negative in case of a beneficial effect, and positive otherwise. In this study, ZDV treatment significantly reduced the factor transfusion requirements of the patients. The observed median sum of changes (range in parentheses) was −964.5 (−3,174 to 1,471) (P = 0.05). The primary pharmacokinetic parameters of CL and V_{ss} were poorly correlated with the pharmacodynamic changes in platelet count (r = −0.07 and −0.53), von Willebrand antigen (r = −0.53 and −0.55), or transfusion requirements (r = 0.47 and 0.75) that were noted during the study.

DISCUSSION

i.v. ZDV administration. Many of the asymptomatic HIV-infected hemophilia patients we examined had decreased total body CLs (~15 ml/min/kg) of ZDV compared with those of AIDS or ARC patients in previous reports. Blum et al. (1), Klecker et al. (15), and deMiranda et al. (6) have

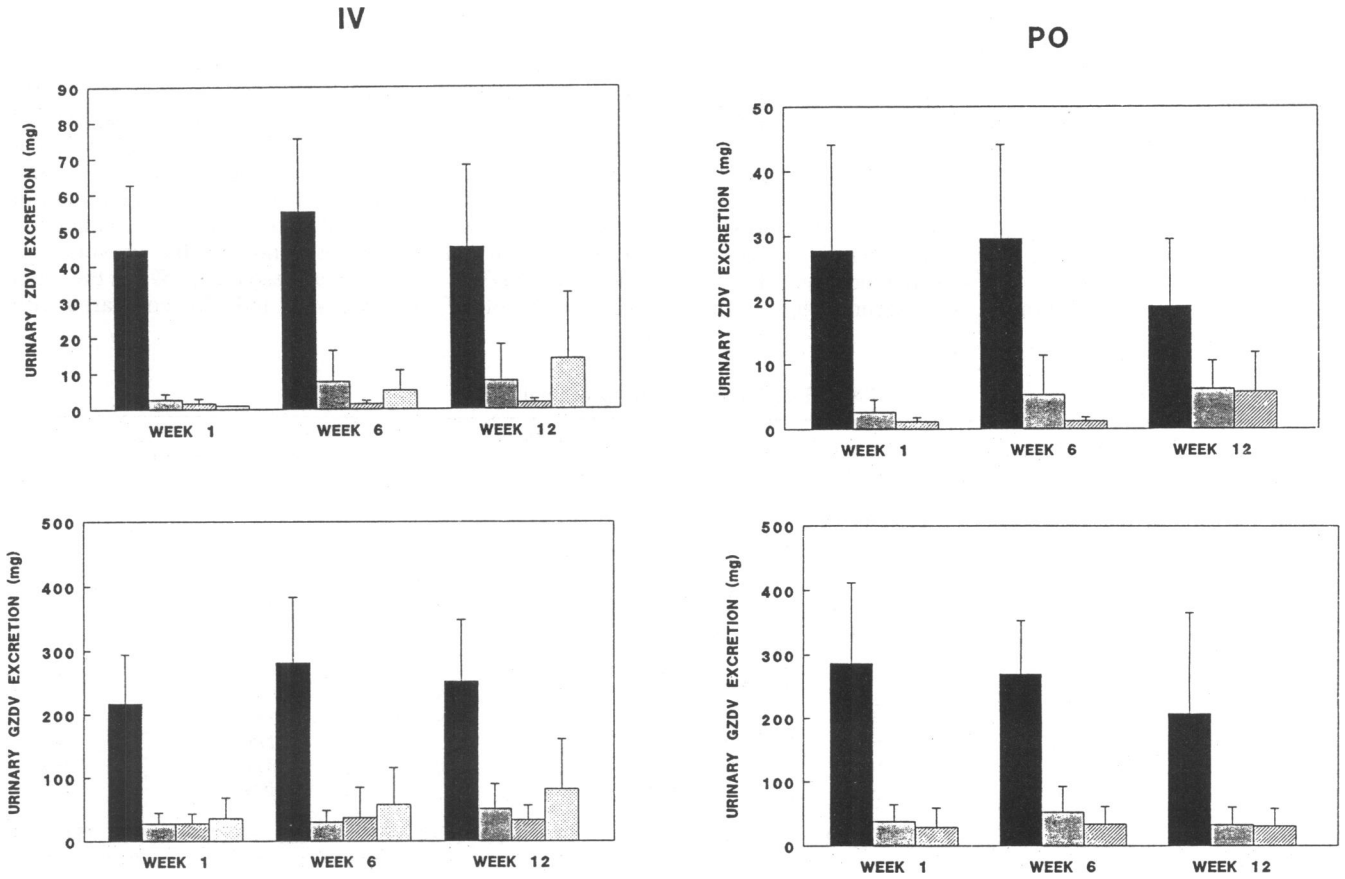

FIG. 2. Urinary recovery patterns (means + standard deviations) of ZDV and GZDV following i.v. and oral (PO) dosing at weeks 1, 6, and 12. The collection intervals were 0 to 4 (■), 4 to 8 (▨), and 8 to 12 (▨) h plus 12 to 24 h (▩) for the i.v. study.

TABLE 4. Pharmacokinetic parameters at weeks 1, 6, and 12 following oral ZDV administration

Patient	Wk 1			Wk 6			Wk 12		
	GZDV peak/ ZDV peak	*F*	CL_{oral} (ml/ min/kg)	GZDV peak/ ZDV peak	*F*	CL_{oral} (ml/ min/kg)	GZDV peak/ ZDV peak	*F*	CL_{oral} (ml/ min/kg)
1	9.68	0.28	26	9.89	0.45	23	9.66	0.57	24
2	4.31	0.99	13	3.14	0.38	34	ND[a]	ND	ND
3	4.14	0.63	23	2.14	0.56	30	2.59	0.51	26
4	2.77	0.54	18	2.88	0.56	14	3.94	0.73	14
5	6.79	0.55	46	2.71	0.47	23	ND	ND	ND
6	4.43	0.26	41	1.56	0.27	37	3.24	0.54	44
7	4.35	0.42	48	3.61	0.15	42	ND	ND	ND
8	1.17	0.64	11	0.99	0.48	18	2.55	0.44	28
9	ND	ND	ND	ND	ND	ND	ND	ND	ND
10	3.69	0.56	34	7.67	0.81	23	4.60	0.77	17
11	ND	ND	ND	ND	ND	ND	ND	ND	ND
Mean ± SD	4.59 ± 2.4	0.54 ± 0.22	29 ± 14	3.95 ± 2.9	0.46 ± 0.19	27 ± 9	4.43 ± 2.7	0.54 ± 0.13	26 ± 11

[a] ND, not determined (see text).

previously reported mean i.v. CL values for adult AIDS or ARC patients receiving dosages of 1 to 5 mg of ZDV per kg of 27.3 ± 3.0, 23.8 ± 4.0, and 35.2 ± 14.1 ml/min/kg, respectively. In addition, Singlas et al. have obtained similar CL values of 36.6 ± 4.7 ml/min/kg with seronegative individuals (28). In addition to the decreased CL among these hemophilia patients, it is noteworthy that the interpatient variability in CL was nearly fivefold (from 5.6 to 26.3 ml/min/kg).

Since most HIV-infected patients are prescribed oral ZDV treatment with a schedule of one dose every 4 h, many recent investigations of ZDV pharmacokinetics have elected to report CL_{oral} during a 4-h study period. However, CL_{oral} determinations may be limited with regard to differentiating among variations in subpopulation patterns of ZDV metabolism or bioavailability. The specific factors associated with decreased ZDV CL in the hemophilia patients remain speculative. All of the hemophilia patients had abnormal hepatic enzyme levels upon entry into the study. This observation is consistent with the presence of chronic hepatitis, a laboratory and pathologic finding that is often observed in the hemophilia patient population (5, 17). Surprisingly, serum albumin, bilirubin, and PT, laboratory values that are usually considered better indicators of metabolic capacity than the presence of hepatitis, were normal in all of the patients. While the metabolism of certain drugs has been shown to be decreased during episodes of acute hepatitis (2, 4), there are no such data available in the hemophiliac population. CL of ZDV was consistent at weeks 6 and 12 in some patients, while in others (patients 1, 2, and 3), CL was lower after 12 weeks. Although hepatic enzyme elevations did vary within and between patients, no correlation between enzyme levels and ZDV CL was noted.

The amounts of ZDV and GZDV recovered in the urine after the first i.v. dose varied between patients. Mass balance calculation with a nanomolar-equivalent correction for GZDV (GZDV × 267.2/443.4 = ZDV in milligrams) yielded a total urinary ZDV recovery of 230 ± 77 mg, an observation that suggests considerable nonrenal, nonhepatic CL (possibly intracellular uptake) in some patients. Interestingly, at week 1, GZDV was recovered in the latter three collection intervals in all patients despite undetectable or very low amounts of ZDV in the serum at these times. When the renal excretion of total ZDV at weeks 6 and 12 is compared with

TABLE 5. Urinary recovery of total ZDV

Patient	Recovery (mg)[a]					
	Wk 1		Wk 6		Wk 12	
	i.v.	Oral	i.v.	Oral	i.v.	Oral
1	186	244	519	309	343	270
2	252	255	349	2,228	226	NC[b]
3	359	319	385	330	465	216
4	251	206	347	305	272	173
5	239	275	250	253	NS[c]	NS
6	138	333	112	292	NS	NS
7	89	241	261	125	99	NS
8	214	254	231	108	184	221
9	296	NS	NS	NS	NS	NS
10	309	NC	337	160	394	389
11	200	NS	NS	NS	NS	NS
Mean ± SD	230 ± 77	266 ± 42	310 ± 114	234 ± 84	283 ± 127	254 ± 83

[a] Calculated as ZDV + (GZDV × 0.6).
[b] NC, urine not collected.
[c] NS, not studied.

TABLE 6. Effect of ZDV on laboratory parameters of coagulation in hemophilia patients

Laboratory parameter (normal range)	No. of patients tested	Median change (range)	P value
Platelet count (160 × 10^3–400 × 10^3/mm^3)	10	44 (−1 to 72)	0.007
Bleeding time (4–8 min)	10	0.5 (−7.8 to 2.5)	0.858
von Willebrand antigen level	9	−80 (−200 to −15)	0.008
Ristocetin cofactor activity (60–150)[a]	9	−40 (−100 to 60)	0.179
PT (10–13)	10	0.05 (−2.8 to 1.8)	0.879
PTT (25–37)	9	16.4 (−51.2 to 45.7)	0.176
Factor VIII activity (60–150)	10	0 (−35 to 10)	0.119
Factor VIII activity (60–150)[b]	6	1 (−0.5 to 7)	0.147
Factor IX activity (60–150)[c]	9	−10 (−40 to 75)	0.636
Fibrinogen level (200–400)	10	−70 (−220 to 172)	0.172

[a] One hemophilia patient with von Willebrand disease was excluded from the group.
[b] One patient with mild hemophilia A and one patient with hemophilia B were excluded from the group.
[c] One patient with hemophilia B was excluded from the group.

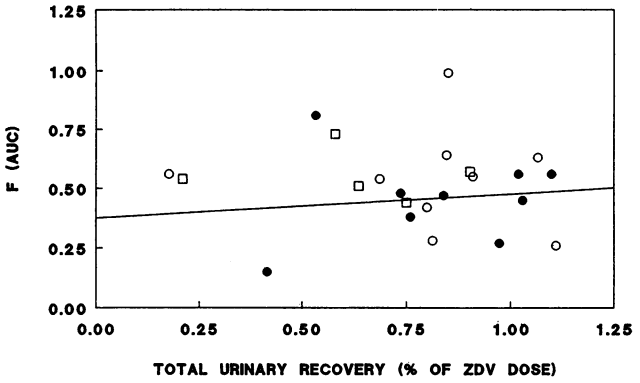

FIG. 3. Correlation of ZDV bioavailability (F) calculated by the ratio of oral to i.v. drug concentrations in plasma [F (AUC)] and total urinary recovery of ZDV [calculated as ZDV + (GZDV × 267.2/442.4)] following i.v. administration (r = 0.12). ○, week 1; ●, week 6; □, week 12.

that at week 1, a variable pattern is present. In the patients studied at week 6, the total urinary recovery increased to 310 ± 114 mg, and at week 12 it was 283 ± 127 mg. Of the seven patients that received an i.v. dose at week 12, five had a decrease from the total recovered at week 6, and two had an increase. These data imply that a sample from a 24-h urine collection from patients that are chronically ingesting ZDV may include drug that was given that day as well as ZDV that is eliminated from a tissue compartment. This theory is supported by the prolonged serum concentration elimination phase seen in many of our patients after the 4-h time point, a pattern suggestive of a deep compartment site of drug distribution.

Oral ZDV administration. Oral ZDV administration in these hemophilia patients has been previously described (20, 21). The i.v. data suggest that previous CL_{oral} values, which are lower than those observed in AIDS or ARC patients, are a result of decreased first-pass metabolism leading to a higher AUC and subsequently to a lower CL_{oral}. Data to support this theory have recently been presented and indicate a lower GZDV-to-ZDV AUC ratio for serum in hemophiliacs than in AIDS or ARC patients (24). Comparison of oral ZDV disposition with i.v. ZDV disposition allows for the determination of bioavailability, and oral ZDV disposition is more indicative of chronic therapy prescribed for HIV-infected individuals. Unfortunately, the F values indicate that both interpatient and intrapatient variability was considerable (Table 3). Indeed, F values increased progressively in some patients, decreased in others, and were consistent in others. No correlation between F values and hepatic enzyme elevations was noted. This would be expected, since Mannucci et al. have previously shown a poor correlation between hepatic enzyme elevation and the degree of biopsy-proven hepatitis (18). Another interesting observation is that there was a poor correlation between bioavailability calculated by $AUC_{oral}/AUC_{i.v.}$ and total urinary recovery of ZDV (ZDV plus GZDV) (Fig. 3). Clearly, the urinary recovery method overestimates the true bioavailability, since values of >1.0 were noted. These data lend further support to the proposed deep tissue compartment for ZDV, which was mentioned above as an explanation for the prolonged plasma elimination phase noted in some of these hemophilia patients.

Coagulation. In this study, ZDV treatment of a small group of HIV+ hemophiliacs did not alter most of the coagulation parameters we measured, including factor VIII, factor VII, factor IX, PT, PTT, fibrinogen level, bleeding time, euglobulin clot lysis time, and presence of factor VIII inhibitor. However, ZDV treatment was associated with a rise in the platelet count and a decline in the von Willebrand antigen level. A rise in platelet count related to ZDV treatment has been noted in several previous studies (11, 12, 19, 25, 29), some including hemophiliacs (22, 23).

The decrease in von Willebrand antigen level associated with ZDV treatment is of uncertain biological significance. Before ZDV treatment, the von Willebrand antigen level was high; it fell to within normal limits only during ZDV administration. Ristocetin cofactor activity also decreased with ZDV treatment, with a median change of −40%, but this change was not statistically significant (P = 0.18). Furthermore, the declines in von Willebrand antigen level and ristocetin cofactor activity did not appear to be related (r_s = 0.250; P = 0.51). A possible explanation for this observation is that while the small multimers of the von Willebrand antigen which are functionally unimportant decreased with ZDV treatment, the large multimers remained essentially unchanged. A report by Budde et al. (3) suggests an alternate and opposite hypothesis. They noted that the decrease or absence of large multimers of the von Willebrand antigen in patients with myeloproliferative disorders or postsplenectomy was directly related to the elevation of the platelet count. They proposed that platelet proteases may cleave von Willebrand antigen or adsorb large von Willebrand antigen onto the platelet membrane. However, in our study, we saw no association between the rise in platelet count and the decrease in von Willebrand antigen (r = 0.212; P = 0.56). Thus, the elevated platelet count seen in our patients during ZDV treatment cannot be used to explain the decrease in von Willebrand antigen.

In summary, the i.v. data obtained in asymptomatic HIV-infected hemophilia patients indicate decreased ZDV CL. Despite the variable ZDV AUC attained within the patients receiving a fixed dose of ZDV, no detrimental effect on coagulation parameters was observed. It should be noted that the recommended dosage of ZDV has been lowered to 500 mg/day since this study was completed. However, the linear pharmacokinetic nature of this drug and the lack of

adverse effects on coagulation parameters at the higher doses that we studied make both the pharmacokinetic and the coagulation data obtained in this study relevant to current ZDV therapy. The long-term administration of ZDV to hemophilia patients will require close clinical monitoring to evaluate the significance of these observed pharmacokinetic alterations. The decreased factor use, the increased platelet counts, and the lack of adverse effect on multiple laboratory parameters of coagulation indicate that HIV^+ hemophilia patients can be treated with ZDV without worsening of their bleeding tendencies.

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