

## Absolute Bioavailability and Metabolic Disposition of Valaciclovir, the L-Valyl Ester of Acyclovir, following Oral Administration to Humans

J. SOUL-LAWTON,\* E. SEABER, N. ON, R. WOOTTON, P. ROLAN,† AND J. POSNER

*Department of Clinical Pharmacology, Wellcome Foundation Ltd., Beckenham, Kent, United Kingdom BR3 3BS*

Received 21 November 1994/Returned for modification 14 March 1995/Accepted 12 July 1995

**Valaciclovir (Valtrex), the L-valyl ester of acyclovir, is undergoing clinical development for the treatment and suppression of herpesviral diseases. The absolute bioavailability of acyclovir from valaciclovir and the metabolic disposition of valaciclovir were investigated with healthy volunteers in two separate studies. In a randomized, crossover study, 12 fasting healthy volunteers each received 1,000 mg of oral valaciclovir and a 1-h intravenous infusion of 350 mg of acyclovir. The mean absolute bioavailability of acyclovir was 54.2%, a value three to five times that obtained previously with oral acyclovir. A similar estimate of 51.3% was made from urinary recovery of acyclovir. In the second study, four fasting volunteers received a single oral dose of 1,000 mg of [<sup>14</sup>C]valaciclovir. The majority of plasma radioactivity was accounted for by acyclovir, with very low plasma valaciclovir concentrations (mean maximum concentration of drug in plasma = 0.19 μM), which were undetectable after 3 h postdose. By 168 h, more than 90% of the administered radioactive dose was recovered, with approximately 46% in urine and 47% in feces. More than 99% of the radioactivity recovered in urine corresponded to acyclovir and its known metabolites, 9-(carboxymethoxymethyl)guanine and 8-hydroxy-9-[(2-hydroxyethoxy)methyl]guanine, with valaciclovir accounting for less than 0.5% of the dose. Acyclovir, but no valaciclovir, was detected in fecal samples. These studies show that after oral administration to humans, valaciclovir is rapidly and virtually completely converted to acyclovir to provide a high level of acyclovir bioavailability in comparison with that following oral administration of acyclovir. The plasma acyclovir exposure obtained following oral administration of valaciclovir is similar to that achieved with doses of intravenous acyclovir, which are effective in the treatment and suppression of the less susceptible herpesviral diseases.**

Valaciclovir (Valtrex), the L-valyl ester of acyclovir, is rapidly and extensively converted to acyclovir after oral administration to humans (13). Acyclovir has *in vitro* inhibitory activity against herpes simplex virus type 1 (HSV-1) and type 2 (HSV-2), varicella-zoster virus, Epstein-Barr virus, and cytomegalovirus (10). The efficacy of acyclovir in the treatment and suppression of HSV-1, HSV-2, and varicella-zoster virus infections is well documented (10), and acyclovir has been shown to play a role in the suppression of cytomegalovirus infections (1, 9, 11). However, the efficacy of oral acyclovir against the less susceptible herpesviruses may be limited by its low oral bioavailability. In humans, the average oral bioavailability after administration of an 800-mg dose is approximately 10% (package insert for Zovirax capsules, tablets, and suspension; Burroughs Wellcome Co., Research Triangle Park, N.C.), increasing to approximately 20% with a 200-mg dose (5). To achieve sufficient plasma acyclovir concentrations for the acute treatment or suppression of less susceptible herpesviral diseases, intravenous therapy may be necessary.

Valaciclovir, after oral administration to animals, was shown to provide significantly higher levels of acyclovir bioavailability than was oral acyclovir (2), and multiple dosing with 1,000 and 2,000 mg of valaciclovir in healthy volunteers provides daily acyclovir area under the plasma concentration time curve (AUC) values approximating those obtained with therapeutic doses of intravenous acyclovir (13).

Metabolic disposition studies with rats and monkeys have indicated that valaciclovir is well absorbed, undergoes extensive presystemic metabolism to acyclovir, and is rapidly distributed and eliminated (4, 6). An enzyme that hydrolyzes valaciclovir to acyclovir, known as valaciclovir hydrolase, has been purified from rat liver mitochondria (3). Acyclovir is further metabolized to the known acyclovir metabolites, 9-(carboxymethoxymethyl)guanine (CMMG) and 8-hydroxy-9-[(2-hydroxyethoxy)methyl]guanine (8-OHACV). The oxidation of acyclovir to CMMG is mediated by the sequential actions of alcohol and aldehyde dehydrogenase, and the 8-hydroxylation of acyclovir is mediated by aldehyde oxidase, pathways which do not involve liver microsomal enzymes (6).

We report here the absolute bioavailability of acyclovir following oral administration of 1,000 mg of valaciclovir and the metabolic disposition of 1,000 mg of [<sup>14</sup>C]valaciclovir when administered to healthy volunteers.

### MATERIALS AND METHODS

**Study design.** Both the absolute-bioavailability and metabolic-disposition studies were carried out with healthy volunteers at the Wellcome Clinical Investigations Unit, King's College Hospital, London, United Kingdom. For entry into either study, volunteers had to have no clinically significant abnormality at the screening medical examination and were excluded if they had participated in other clinical trials or donated blood during the preceding 2 to 3 months, showed the presence of drugs of addiction in the urine, or had a history of intolerance to acyclovir. All subjects included in the study gave written informed consent before participation. Prior to the start of the study, the protocols were approved by the Wellcome Protocol Review Committee and the hospital Ethics Committee, and approval from the Administration of Radioactive Substance Advisory Committee (ARSAC) to permit the administration of the radioactive substance to humans was obtained.

**(i) Absolute bioavailability.** Twelve healthy volunteers of either sex, aged 18 to 55 years, were eligible to participate in the open, randomized, 2-period crossover study to determine absolute bioavailability. Volunteers were excluded if they

\* Corresponding author. Phone: 0181 658 2211 (ext. 26310). Fax: 0181 663 0509.

† Present address: Medeval Limited, University of Manchester, Skelton House, Manchester Science Park, Manchester, United Kingdom M15 6SH.

were taking drugs other than an oral contraceptive or, if female, they were pregnant or lactating or, if of child-bearing potential, they were not employing adequate contraceptive measures. Smoking and the consumption of caffeine-containing beverages were not allowed on the study day. After an overnight fast, volunteers received, on separate occasions, a single dose of 1,000 mg of valaciclovir orally or 350 mg of acyclovir as a 1-h intravenous infusion with an interval of at least 1 week between doses. The dose and duration of infusion of acyclovir were chosen to produce a plasma concentration-time profile similar to that following administration of 1,000 mg of valaciclovir, assuming approximately 50% bioavailability of acyclovir from valaciclovir and taking into account the amount of releasable acyclovir in valaciclovir (13). Blood samples were taken before dosing and then 15, 30, 45, and 60 min and 1.5, 2.0, 2.5, 3.0, 4.0, 5.0, 6.0, 8.0, 10, 12, 16, and 24 h following each dose, and the plasma was separated and frozen at  $-20^{\circ}\text{C}$  for assay of acyclovir. All urine was collected, in four fractions up to 24 h. The weight of each fraction was recorded and used as the urinary volume, and after mixing, a 10-ml aliquot was taken and frozen at  $-20^{\circ}\text{C}$  for assay of acyclovir.

**(ii) Metabolic disposition.** Four healthy volunteers of either sex, aged 30 to 55 years, were eligible to participate in the metabolic-disposition study, and they received a single oral dose of [ $^{14}\text{C}$ ]valaciclovir on one occasion only. Females had to be incapable of child bearing (postmenopausal, hysterectomy, or sterilized), and males had to give an undertaking that they and/or their partner would use an adequate and reliable form of contraception for not less than 3 months from the first day of the study. Volunteers were excluded if they were taking other medication. After an overnight fast, volunteers received 1,000 mg of [ $^{14}\text{C}$ ]valaciclovir (50  $\mu\text{Ci}$ ). Blood samples were taken before dosing and at 0.5, 1.0, 1.5, 2.0, 2.5, 3.0, 4.0, 6.0, 8.0, 10, 12, 15, 24, 32, 48, 72, and 96 h postdose. All urine was collected, in seven fractions up to 96 h, and the weight of each fraction was recorded as described above. All stools were collected up to 168 h (7 days) postdose. At all time points, a 1-ml sample of whole blood was added to 4 ml of distilled water to form a hemolysate and stored at  $-20^{\circ}\text{C}$  for total radioactivity determination. The remaining blood was kept on ice. Blood samples were centrifuged at  $4^{\circ}\text{C}$ , and the plasma was frozen in aliquots at  $-20^{\circ}\text{C}$  for determination of total radioactivity, radioimmunoassay for acyclovir, and high-performance liquid chromatography (HPLC) assay for valaciclovir. Valaciclovir was assayed only up to 3 h postdose. Aliquots of urine were taken for total radioactivity determination and acyclovir assay. For samples containing sufficient radioactivity, HPLC profiling was carried out for the simultaneous analysis of acyclovir and valaciclovir. Stool samples were homogenized in water, and the homogenate was weighed. Total radioactivity in an aliquot of the homogenate was then determined, and HPLC profiling was carried out with supernatants obtained from centrifugation of the fecal homogenates as described for urine.

**Study drugs.** All drugs were supplied by the Wellcome Foundation Ltd., and tablets and capsules were taken with 200 ml of water.

**(i) Absolute-bioavailability study.** Oral valaciclovir was administered as white, film-coated 500-mg tablets. Acyclovir for intravenous infusion (Zovirax) was provided in vials containing the sodium salt equivalent to 500 mg of acyclovir. This was reconstituted with 0.9% sodium chloride for injection, and 350 mg of acyclovir was infused over a 1-h period via an IVAC infusion pump.

**(ii) Metabolic-disposition study.** Oral valaciclovir was administered as opaque capsules containing 125 mg of valaciclovir with a specific activity of 0.05  $\mu\text{Ci}/\text{mg}$ .

**Safety.** Any adverse experiences (AEs) occurring during the study period were recorded. Blood samples for hematological and biochemical parameter estimations were taken from each subject at baseline and at the end of the study period. These data were inspected for clinically significant abnormalities or changes.

**Plasma and urine assays for acyclovir and valaciclovir.** Valaciclovir concentrations in plasma were determined by using a validated, specific, reversed-phase HPLC method described previously (13). The standard curve was linear over the concentration range from 0.25 to 50  $\mu\text{M}$ . The lower limit of quantitation for valaciclovir was 0.25  $\mu\text{M}$ , with precision shown by coefficients of variation less than 5%. Plasma and urine acyclovir determinations were made by using double-antibody radioimmunoassay, which utilizes a monoclonal antibody raised to acyclovir and [ $^3\text{H}$ ]acyclovir as a tracer. This method separates antibody-bound from free [ $^3\text{H}$ ]acyclovir and is a modification of the original radioimmunoassay method for acyclovir assay (12). The standard curve was linear over the concentration range from 0.03 to 0.5  $\mu\text{M}$ . The lower limit of quantitation of the assay was 0.05  $\mu\text{M}$  for plasma and 0.8  $\mu\text{M}$  for urine, with precision shown by coefficients of variation of 10% or less. Acyclovir is stable in frozen human plasma at  $-20^{\circ}\text{C}$  for at least 12 months, though samples were analyzed within 4 months of being taken.

**Determination of radioactivity.** Radioactivity in plasma and urine was determined directly by liquid scintillation counting (LSC) with a Beckman scintillation counter by using an external standard for quench correction. Measured aliquots of plasma or urine were weighed directly into scintillation vials and mixed with scintillator cocktail. Counting efficiencies were  $>80$  and  $>90\%$  for plasma and urine, respectively. For hemolysate, measured aliquots from each sample were oxidized prior to LSC. For feces, a homogenate in water was prepared from each collection and weighed aliquots were oxidized prior to LSC. Supernatants were obtained by centrifugation of the fecal homogenates for HPLC profiling.

**Radiochromatography.** All urine and fecal samples which contained greater than 10% of the administered radioactive dose were analyzed by HPLC by using a Milton Roy/LDC HPLC system with CM3000 pumps, a GM 4000 gradient

programmer, and an SM 4000 UV detector (at 245 nm), coupled to a Berthold LB506C-1 radiochemical detector. Both the UV and radiochemical data were captured and processed by using the Berthold data system. A Hewlett-Packard HP1090 liquid chromatograph coupled to a PE-Sciex API-III mass spectrometer via a fully articulated Ionspray (ISP) source was used for analysis by liquid chromatography-mass spectrometry. Urine was filtered through a 0.8- $\mu\text{m}$ -pore-size Millex-AA filter unit (Millipore) and fecal homogenates were centrifuged before being stabilized with 10% (wt/vol) trichloroacetic acid, and both were injected directly (100  $\mu\text{l}$ ) onto the column.

HPLC analysis was performed with a Spherisorb S5-ODS1 column (inside diameter, 25 cm by 4.6 mm) at ambient temperature. The mobile phase was 25 mM ammonium formate buffer, pH 3.5 (buffer A), and 50 mM ammonium acetate buffer, pH 5.5: acetonitrile, 1:1 (vol/vol) (buffer B). The solvent gradient program used was 0 to 10% buffer B in buffer A over 30 min and then 10 to 100% buffer B in buffer A over 15 min, after which 100% buffer B was maintained for a further 10 min. A flow rate of 1.0 ml/min was used throughout. Column recoveries were  $>84\%$  for urine and  $>90\%$  for feces.

For analysis by liquid chromatography-mass spectrometry, an HPLC system similar to that used for radioprofiling was used and the eluate was split such that approximately 30  $\mu\text{l}/\text{min}$  entered the ion source. Argon was used as the collision gas during tandem mass spectrometry.

**Pharmacokinetic and statistical analysis. (i) Absolute-bioavailability study.** Noncompartmental pharmacokinetic parameters were determined for plasma acyclovir following oral administration of valaciclovir and intravenous administration of acyclovir. The observed peak concentration of drug in plasma ( $C_{\text{max}}$ ) and the time to reach the peak concentration ( $T_{\text{max}}$ ) were taken directly from the plasma concentration-time profiles. The AUC from zero to the last measurable concentration ( $\text{AUC}_{0-t}$ ) was estimated by the linear trapezoidal rule. The AUC from zero to infinity ( $\text{AUC}_{0-\text{inf}}$ ) was calculated as  $\text{AUC}_{0-\text{inf}} = \text{AUC}_{0-t} + C_t/\lambda_z$ , where  $C_t$  is the last quantifiable concentration at time  $t$  and  $\lambda_z$  is the first-order elimination rate constant.  $\lambda_z$  was obtained by log linear regression by using five datum points between 6 and 16 h postdose.

The absolute bioavailability ( $F$ ) of acyclovir from valaciclovir was calculated from  $F = \text{AUC}_{\text{p.o.}}/\text{AUC}_{\text{i.v.}} \times \text{dose}_{\text{i.v.}}/\text{dose}_{\text{p.o.}}$  (molar units), where p.o. is oral and i.v. is intravenous.

The elimination half-life ( $t_{1/2}$ ) of acyclovir was calculated as  $t_{1/2} = \ln 2/\lambda_z$ .

Clearance of acyclovir following intravenous administration (CL) was calculated as  $\text{CL} = \text{dose}/\text{AUC}_{0-\text{inf}}$ .

Urinary recovery was calculated as  $\text{Ae}_{0-24}/\text{dose}$ , where Ae is the amount excreted in urine over 24 h, and the ratio of the mean 24-h urinary recovery of acyclovir following administration of valaciclovir and intravenous acyclovir was also calculated to estimate the systemic bioavailability of acyclovir from oral valaciclovir. Renal clearance of acyclovir ( $\text{CL}_{\text{R}}$ ) was calculated as  $\text{CL}_{\text{R}} = \text{Ae}_{0-24}/\text{AUC}_{0-\text{inf}}$ .

The statistical differences between the pharmacokinetic parameters  $\text{AUC}_{0-\text{inf}}$  and  $t_{1/2}$  obtained with the two formulations were subjected to analysis of variance, taking into account variation due to subject, period, and treatment. Dose-adjusted AUC values were log transformed prior to analysis. Data were back transformed to provide a point estimate with 95% confidence intervals of the absolute bioavailability of acyclovir from valaciclovir, and for the difference between  $t_{1/2}$  values following oral administration of valaciclovir and intravenous administration of acyclovir.

**(ii) Metabolic-disposition study.** From plasma valaciclovir data,  $C_{\text{max}}$ ,  $T_{\text{max}}$ , and  $\text{AUC}_{0-t}$  only were determined. From plasma acyclovir data,  $C_{\text{max}}$ ,  $T_{\text{max}}$ ,  $\text{AUC}_{0-\text{inf}}$ , and  $t_{1/2}$  were determined. These pharmacokinetic parameters and  $\text{CL}_{\text{R}}$  were calculated as described above for the absolute-bioavailability study.  $C_{\text{max}}$ ,  $T_{\text{max}}$ , and AUC were calculated for plasma radioactivity levels expressed as micromole equivalents. Percent recoveries of individual compounds and total radioactivity in urine and feces were calculated relative to the calculated dose of valaciclovir in micromoles and microcuries.

No formal statistical analysis was carried out for data for the four subjects in this study, and results are presented in tabular form.

## RESULTS

**Subjects.** For the absolute-bioavailability study, 12 healthy volunteers (4 males and 8 females) with a mean age of 36 years (range, 23 to 50 years) were enrolled. Mean height was 1.68 m (range, 1.53 to 1.85 m), and mean weight was 73 kg (range, 56 to 114 kg). For the metabolic-disposition study, four healthy volunteers (two males and two females) with a mean age of 48 years (range, 36 to 53 years) were enrolled. Mean height was 1.63 m (1.51 to 1.74 m), and mean weight was 74 kg (range, 62 to 83 kg). All subjects enrolled completed the studies, and there were no major deviations from the protocol.

**Pharmacokinetics. (i) Absolute bioavailability.** The mean plasma acyclovir concentration-time profiles following administration of intravenous acyclovir and oral valaciclovir are

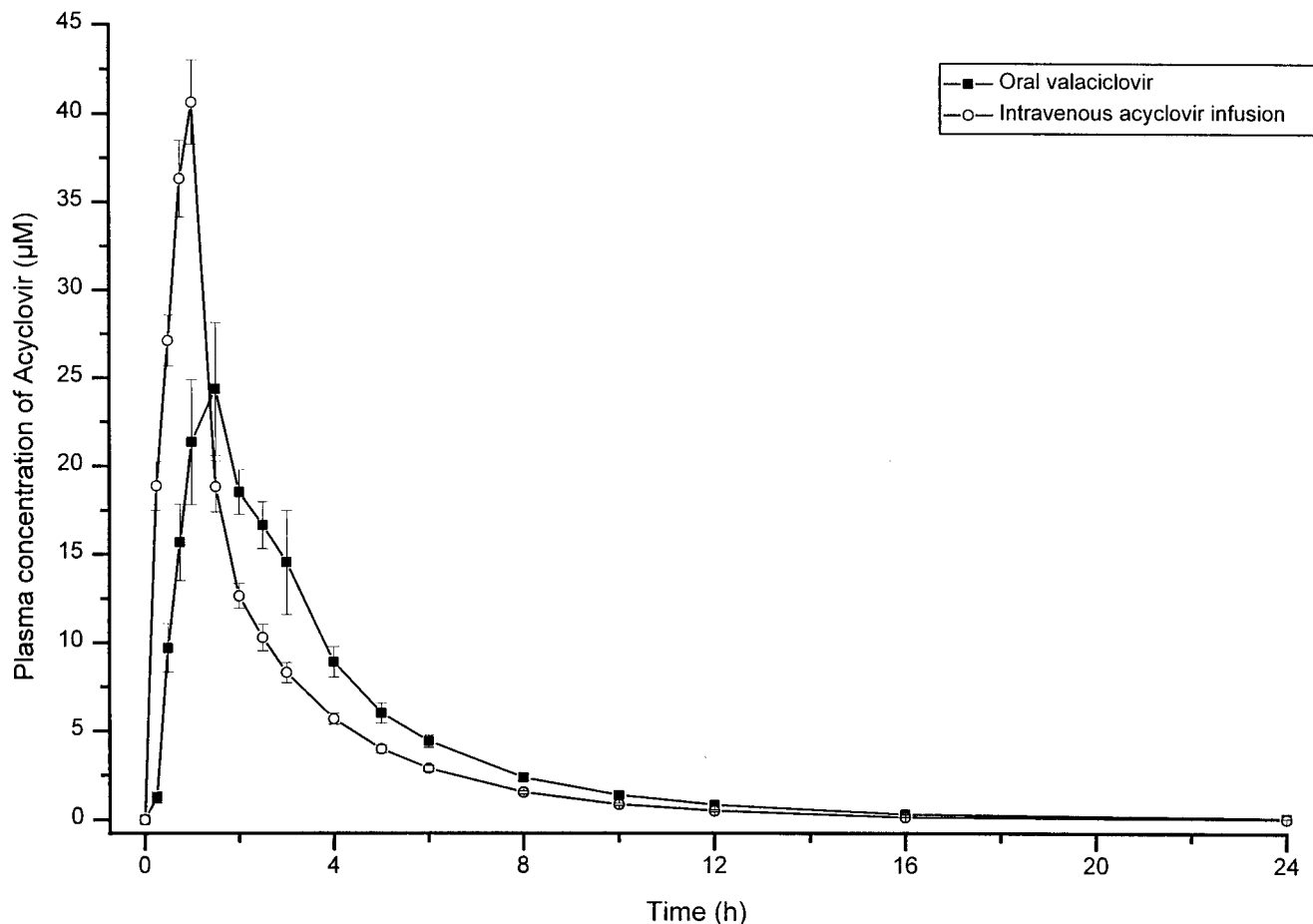


FIG. 1. Mean plasma concentration-time profiles of acyclovir following administration of 1,000 mg of oral valaciclovir or a 350-mg intravenous infusion of acyclovir over 1 h.

shown in Fig. 1, and acyclovir pharmacokinetic parameters derived from plasma drug concentration and urinary recovery data are given in Table 1. Compared with values for oral valaciclovir, the mean acyclovir  $C_{max}$  was higher and the  $T_{max}$  was shorter following administration of intravenous acyclovir. Comparable values for AUC (84.0 and 89.4  $\mu\text{M} \cdot \text{h}$ ) and  $\text{CL}_R$  (283 and 255 ml/min) were obtained for the two formulations (intravenous acyclovir and oral valaciclovir, respectively).

The absolute bioavailability of acyclovir from oral valaciclovir determined from plasma data was 54.2% (95% confidence intervals, 49.4 and 59.5); the range of values was 42 to 73%. A similar estimate of 51.3% was calculated from the ratio of urinary recovery of acyclovir following treatment with oral valaciclovir and intravenous acyclovir. A mean difference of 0.25 h in  $t_{1/2}$  was statistically significantly different for acyclovir for the two formulations.

**(ii) Metabolic disposition.** The plasma profiles of mean valaciclovir and acyclovir concentrations up to 32 h postdose are given in Fig. 2. Valaciclovir was undetectable after 3 h in all subjects and acyclovir was undetectable by 96 h. The sum of plasma valaciclovir and acyclovir profiles and plasma radioactivity profiles are shown in Fig. 3. Radioactivity was undetectable by 8 h postdose. The profiles are similar in shape, but the concentrations of radioactivity in plasma are slightly higher at all time points than the summed acyclovir and valaciclovir concentrations except where the radioactivity falls below the level of quantification because of the lesser sensitivity of radioactivity measurement than of measurement by radioimmunoassay. Radioactivity in blood hemolysate was less than the limit of detection, except for one sample for one subject at 1.5 h postdose, with a value of 0.005  $\mu\text{M}$  equivalents per g.

Noncompartmental pharmacokinetic parameters for plasma

TABLE 1. Acyclovir pharmacokinetic parameters (mean  $\pm$  SD) following administration of 1,000 mg of oral valaciclovir or 350 mg of acyclovir as a 1-h infusion

Administered drug	$C_{max}$ ( $\mu\text{M}$ )	$T_{max}$ (h)	AUC ( $\mu\text{M} \cdot \text{h}$ )	$t_{1/2}$ (h)	CL (ml/min)	$\text{CL}_R$ (ml/min)	% Urinary recovery
Oral valaciclovir	29.53 $\pm$ 12.47	1.71 $\pm$ 0.69	89.37 $\pm$ 19.37	2.62 $\pm$ 0.35	NA <sup>a</sup>	255.3 $\pm$ 85.8	45.8 $\pm$ 17.1
Intravenous acyclovir	40.99 $\pm$ 8.35	0.98 $\pm$ 0.07	84.04 $\pm$ 14.16	2.37 $\pm$ 0.31	322.0 $\pm$ 68.0	282.9 $\pm$ 103.0	86.7 $\pm$ 19.9

<sup>a</sup> NA, not applicable.

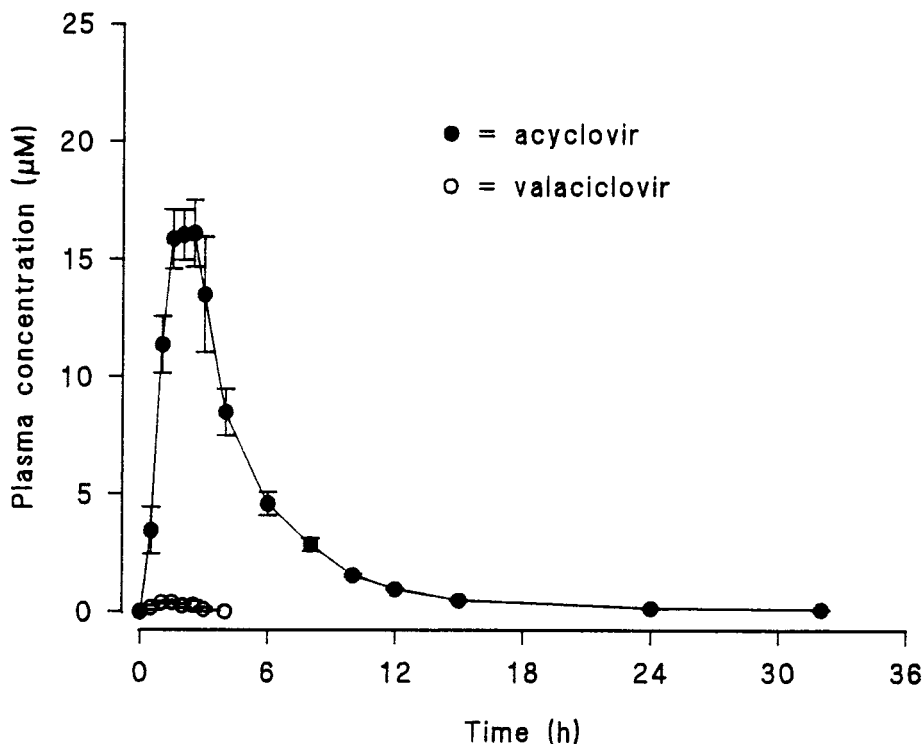


FIG. 2. Mean plasma acyclovir and valaciclovir profiles after administration of 1,000 mg of [ $^{14}\text{C}$ ]valaciclovir to four healthy volunteers.

valaciclovir, acyclovir, and radioactivity are given in Table 2. The mean AUC values for acyclovir and plasma radioactivity were similar at  $80.7 \mu\text{M} \cdot \text{h}$  and  $82.9 \mu\text{M}$  equivalents  $\cdot \text{h}$ , while the AUC value for valaciclovir was only  $0.81 \mu\text{M} \cdot \text{h}$ . The mean  $T_{\text{max}}$  values for acyclovir and plasma radioactivity were almost identical at 2.25 and 2.30 h, respectively, while the valaciclovir  $T_{\text{max}}$  was 1.5 h.  $C_{\text{max}}$  for valaciclovir was approximately only 3% that of acyclovir. The  $t_{1/2}$  values for plasma radioactivity and acyclovir were 1.9 and 2.7 h, respectively. The difference in  $t_{1/2}$  is likely to be due to differences in assay sensitivity, resulting in  $t_{1/2}$  estimates being calculated from different portions of the elimination curve. For acyclovir,  $t_{1/2}$  was estimated from the points between 6 and 15 h, and for total radioactivity,  $t_{1/2}$  was estimated from the points between 2 and 6 h.

By 168 h postdose, 92.7% (range, 84.8 to 101.1%) of the administered radioactive dose had been recovered, with 45.6% (range, 33.4 to 52.8%) in urine and 47.1% (range, 36.8 to 54.9%) in feces. The mean  $\pm$  standard deviation (SD) for total urinary recovery of acyclovir was  $40.4\% \pm 10.8\%$ . The mean difference between the urinary recoveries of acyclovir and radioactivity was 5.2%.

Radiochromatographic HPLC analysis of urine samples confirmed that the majority of the recovered radioactivity was acyclovir, constituting  $84.39\% \pm 4.68\%$  of the total material recovered from urine collections containing  $>10\%$  of the administered dose over the 0- to 8-h postdose period (means and SDs were calculated for three subjects only as there was insufficient radioactivity detected in the 4- to 8-h postdose period for analysis by HPLC for one subject). A peak which cochromatographed with the known acyclovir metabolite CMMG accounted for  $13.78\% \pm 4.2\%$  of the radioactivity recovered up to 8 h postdose. A further metabolite which cochromatographed with another known, minor metabolite of acyclovir, 8-OHACV, accounted for  $1.46\% \pm 1.06\%$  of the recovered

radioactivity up to 8 h postdose. Thus, acyclovir and the known acyclovir metabolites, CMMG and 8-OHACV, accounted for most of the radioactivity. Valaciclovir was detected in only one urine sample (0 to 4 h) from one subject and accounted for less than 0.5% of the total dose.

Radiochromatographic HPLC analysis was carried out with fecal samples from the 24- to 48-h postdose collection period only. No valaciclovir was detected in any of the fecal samples analyzed, acyclovir only was detected in two of the three samples analyzed, and 8-OHACV was detected in a third sample but accounted for only 1.5% of the administered dose.

**AEs.** Eight subjects in the absolute-bioavailability study and one subject in the metabolic-disposition study reported AEs, none of which was serious. The majority of the AEs were headache (six of nine subjects), and in one subject only, in the absolute-bioavailability study, was an AE (headache) considered possibly related to study medication, following 1,000 mg of valaciclovir. There were no clinically significant changes in hematology or biochemistry parameters during the study.

## DISCUSSION

Results of the two studies reported here agree with previous findings with humans that valaciclovir is rapidly and extensively converted to acyclovir and known acyclovir metabolites after oral administration (13).

The absolute bioavailability of 54% for acyclovir is consistent with values obtained with animals (4, 6) and confirms that, in humans, the bioavailability of acyclovir from oral valaciclovir is three to five times greater than that achieved following treatment with oral acyclovir. The small but statistically significant increase in  $t_{1/2}$  following treatment with valaciclovir compared with intravenous acyclovir is not clinically significant and

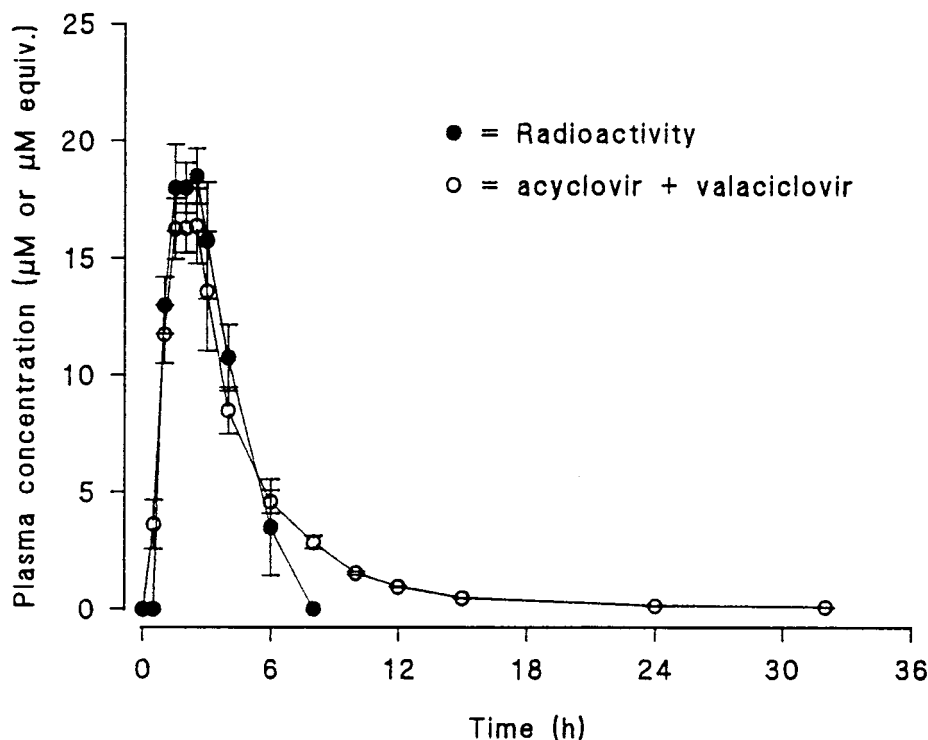


FIG. 3. Mean plasma radioactivity and summed acyclovir plus valaciclovir concentrations following administration of 1,000 mg of [ $^{14}\text{C}$ ]valaciclovir to four healthy volunteers.

is probably due to a small component of continuing absorption of valaciclovir.

In all subjects in the radiolabel study, the sum of acyclovir and valaciclovir concentrations was slightly lower than the corresponding level of plasma radioactivity, suggesting the presence of low concentrations of circulating metabolite(s). Similarly, the mean recovery of acyclovir in urine was lower than that of radioactivity, indicating the presence of metabolites in urine. The identity and proportions of metabolites observed in the metabolic-disposition study with [ $^{14}\text{C}$ ]valaciclovir are similar to those obtained in a previous study following administration of intravenous [ $^{14}\text{C}$ ]acyclovir (7). In the latter study, CMMG was the only significant urinary metabolite, accounting for 8.5 to 14.1% of the administered dose, and 8-OHACV accounted for less than 0.5% of the dose. The comparable recoveries of metabolites in the present study, together with the fact that negligible valaciclovir was detected in the urine, indicate that valaciclovir is fully converted to acyclovir after absorption and levels of CMMG and 8-OHACV observed are due to further metabolism of acyclovir. No valaciclovir was detected in stool samples; they contained only acyclovir, with the exception of one sample in which a small amount of

8-OHACV was detected. This indicates that, in addition to absorbed valaciclovir being converted to acyclovir, unabsorbed valaciclovir is also converted to acyclovir within the gut lumen; this may explain the less than 100% bioavailability of acyclovir following oral dosing with valaciclovir in humans.

The unquantifiable levels of radioactivity in hemolysate samples after a fivefold dilution in water suggest that acyclovir is not concentrated inside erythrocytes and agrees with a previous study in which [ $^{14}\text{C}$ ]acyclovir was administered and approximately equivalent levels of radioactivity were found in blood and plasma (7).

The 1000-mg dose of valaciclovir used in the two studies was well tolerated by all the healthy volunteers. This is consistent with the good safety profile observed in previous phase 1 studies with valaciclovir in healthy volunteers (13) and patients with human immunodeficiency virus infection (8).

In summary, when valaciclovir, the L-valyl ester of acyclovir, was orally administered to humans, approximately 54% of the dose was absorbed. Of the absorbed valaciclovir, more than 99% was rapidly converted to acyclovir to give high plasma acyclovir concentrations and low plasma valaciclovir concentrations which became undetectable after 3 h postdose. No

TABLE 2. Pharmacokinetic parameters (mean  $\pm$  SD) for valaciclovir and acyclovir and from plasma radioactivity following administration of 1,000 mg of [ $^{14}\text{C}$ ]valaciclovir to four healthy volunteers

Drug or parameter	$C_{\max}$ ( $\mu\text{M}$ )	$T_{\max}$ (h)	AUC ( $\mu\text{M}\cdot\text{h}$ )	$t_{1/2}$ (h)
Valaciclovir	$0.59 \pm 0.17$	$1.50 \pm 0.71$	$0.81 \pm 0.26$	NQ <sup>a</sup>
Acyclovir	$18.69 \pm 1.34$	$2.25 \pm 0.65$	$80.69 \pm 9.27$	$2.74 \pm 0.21$
Plasma radioactivity	$21.00 \pm 1.80^b$	$2.30 \pm 0.65$	$82.90 \pm 13.50^c$	$1.93 \pm 0.45$

<sup>a</sup> NQ, not quantifiable.

<sup>b</sup> Units are micromolar equivalents.

<sup>c</sup> Units are micromolar-per-hour equivalents.

unknown metabolites were detected, with acyclovir and known acyclovir metabolites, CMMG and 8-OHACV, accounting for the absorbed dose. Following oral administration, valaciclovir provides plasma acyclovir exposure similar to that achieved with doses of intravenous acyclovir which are effective in the treatment and suppression of the less susceptible herpesviral diseases.

#### ACKNOWLEDGMENTS

We acknowledge the contributions of Elaine Tate, Lewis Kanics, and Anne Nicholls for bioanalytical support.

#### REFERENCES

1. Balfour, H. H., B. A. Chace, J. T. Stapleton, R. L. Simmons, and D. S. Fryd. 1989. A randomized placebo-controlled trial of oral acyclovir for the prevention of cytomegalovirus disease in recipients of renal allografts. *N. Engl. J. Med.* **320**:1381–1387.
2. Beauchamp, L. M., G. F. Orr, P. de Miranda, T. Burnette, and T. A. Krenitsky. 1992. Amino acid ester prodrugs of acyclovir. *Antivir. Chem. Chemother.* **3**(3):157–164.
3. Burnette, T. C., and P. de Miranda. 1993. Purification and characterization of an enzyme from rat liver that hydrolyzes 256U87, the L-valyl ester prodrug of acyclovir (Zovirax<sup>®</sup>). *Antivir. Res.* **20**(Suppl. 1):115.
4. Burnette, T. C., and P. de Miranda. 1994. Metabolic disposition of the acyclovir prodrug valaciclovir in the rat. *Drug Metab. Dispos.* **22**:60–64.
5. de Miranda, P., and M. R. Blum. 1983. Pharmacokinetics of acyclovir after intravenous and oral administration. *J. Antimicrob. Chemother.* **12**(Suppl. B):29–37.
6. de Miranda, P., and T. C. Burnette. 1994. Metabolic fate and pharmacokinetics of the acyclovir prodrug valaciclovir in cynomolgus monkeys. *Drug Metab. Dispos.* **22**:55–59.
7. de Miranda, P., S. S. Good, O. L. Laskin, H. C. Krasny, J. D. Connor, and P. S. Lietman. 1981. Disposition of intravenous radioactive acyclovir. *Clin. Pharmacol. Ther.* **30**:662–672.
8. Jacobson, M. A., J. Gallant, L. Wang, D. Coakley, S. Weller, D. Gary, L. Squires, M. L. Smiley, M. R. Blum, and J. Feinberg. 1994. Phase 1 trial of valaciclovir, the L-valyl ester of acyclovir, in patients with advanced human immunodeficiency virus disease. *Antimicrob. Agents Chemother.* **38**:1534–1540.
9. Meyers, J. D., E. C. Reed, D. H. Shepp, M. Thornquist, P. S. Dandliker, C. A. Vicary, N. Flournoy, L. E. Kirk, J. H. Kersey, E. D. Thomas, and H. H. Balfour. 1988. Acyclovir for prevention of cytomegalovirus infection and disease after allogeneic marrow transplantation. *N. Engl. J. Med.* **318**:70–75.
10. O'Brien, J. J., and D. M. Campoli-Richards. 1989. Acyclovir. An updated review of its antiviral activity, pharmacokinetic properties and therapeutic efficacy. *Drugs* **37**:233–309.
11. Prentice, H. G., E. Gluckman, R. C. Powles, P. Ljungman, N. J. Milpied, J. M. F. Ranada, F. Madelli, P. Kho, L. Kennedy, and A. R. Bell. 1994. Impact of long-term acyclovir on cytomegalovirus infection and survival after allogeneic bone marrow transplantation. *Lancet* **343**:749–753.
12. Quinn, R. P., P. de Miranda, L. Gerald, and S. S. Good. 1979. A sensitive radioimmunoassay for the antiviral agent BW248U [9-(2-hydroxyethoxymethyl)guanine]. *Anal. Biochem.* **98**:319–328.
13. Weller, S., M. R. Blum, M. Doucette, T. Burnette, D. M. Cederberg, P. de Miranda, and M. L. Smiley. 1993. Pharmacokinetics of the acyclovir prodrug valaciclovir after escalating single- and multiple-dose administration to normal volunteers. *Clin. Pharmacol. Ther.* **54**:595–605.