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Effects of Probenecid on the Pharmacokinetics and Elimination of Acyclovir in Humans

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The effects of probenecid on the pharmacokinetics and renal clearance of acyclovir were studied in humans. Acyclovir (5 mg/kg) was given as a 1-h infusion to three volunteers with normal renal function both before and after oral administration of probenecid (1 g). The kinetics were well described by a two-compartment open model with zero-order infusion. The mean acyclovir concentrations at all time points after 1.0 h from the end of acyclovir infusion following probenecid administration were statistically higher than the corresponding mean acyclovir concentrations following the acyclovir infusion without probenecid administration. In the absence of probenecid, the renal clearance (248 ± 80 ml/min per 1.73 m^2) accounted for 83% of the total clearance (300 ± 69 ml/min per 1.73 m^2) and was almost threefold greater than the estimated creatinine clearance (90 ± 48 ml/min per 1.73 m^2). After probenecid administration, there was a 32% decline in renal clearance (248 to 168 ml/min per 1.73 m^2; $P \leq 0.05$), a 40% increase in the area under the curve (91.3 to 127.6 nmol.h/ml; $P < 0.05$), and an 18% increase in the terminal plasma half-life (2.3 to 2.7 h; $P < 0.01$). Although statistically significant, these effects due to the influence of probenecid probably have only limited clinical importance. In this study we confirmed that acyclovir is eliminated predominantly by renal clearance, both by glomerular filtration and tubular secretion; our results suggested that at least part of the tubular secretion is inhibited by probenecid.

Acyclovir, a potent anti-herpesvirus agent, is eliminated predominantly by renal excretion (2, 4, 5, 7, 13). The only significant metabolite of acyclovir which has been isolated to date is 9-carboxymethoxymethylguanine (4, 5), which accounts for less than 14% of the administered dose. The renal clearance of acyclovir after a single 1-h infusion is two- to threefold greater than the creatinine clearance (4, 5, 7, 13). This suggests that acyclovir is eliminated by other renal mechanisms in addition to glomerular filtration and that renal tubular secretion may play an important role in acyclovir elimination. The mechanism of this tubular secretion is not known.

Probenecid, an inhibitor of the organic acid transport system (1, 3), has been used frequently to elucidate the role of organic acid transport in the fate of various drugs (3). We conducted a study in human volunteers to evaluate the effects of probenecid on the pharmacokinetics and elimination of acyclovir.

MATERIALS and METHODS

Patient selection. Three patients (two females and one male) with a mean age of 47.3 years (range, 24 to 67 years) were selected. Each of these patients had a malignancy; two had a hematological malignancy, and one had carcinoma of the breast. The mean weight, height, and body surface area of these patients were 79.1 kg (range, 68 to 100 kg), 159 cm (range, 155.1 to 164 cm), and 1.89 m^2 (range, 1.75 to 2.17), respectively. The prognosis for each patient was such that the life expectancy was at least 6 months, and in each case the long-term prognosis was good. Informed consent was obtained from each subject before entry into the study. None of the subjects had either renal or hepatic dysfunction, was receiving pyrimidine or purine analogs or other drugs which affected purine metabolism or excretion, had received any antiviral therapy in the prior week, or was receiving concurrent maintenance therapy for his or her malignancy.

Drug, administration, and dosage. Acyclovir was provided by the Burroughs Wellcome Co. as the sterile, lyophilized sodium salt. Each 50-ml vial, containing 500 mg of acyclovir as the sodium salt, was reconstituted with 25 ml of sterile water for injection.

TABLE 1. Mean plasma acyclovir concentrations in the absence and presence of probenecid

Time (h)[a]	Acyclovir concn (μg/ml)	
	In the absence of probenecid	In the presence of probenecid
0.25	4.5 ± 0.35[b]	4.05 ± 0.35
0.5	6.35 ± 0.19	6.99 ± 0.84
0.75	7.41 ± 0.44	8.67 ± 1.05
1.0	11.4 ± 2.42	11.3 ± 0.79
1.5	4.80 ± 0.51	5.99 ± 0.79
2.0	3.08 ± 0.27	4.68 ± 0.68
3.0	2.16 ± 0.39	3.29 ± 0.41
4.0	1.39 ± 0.33	2.21 ± 0.35
6.0	0.79 ± 0.21	1.41 ± 0.32
8.0	0.47 ± 0.13	0.85 ± 0.14
10.0	0.27 ± 0.07	0.52 ± 0.10
12.0	0.14 ± 0.05	0.31 ± 0.07

[a] Time from the start of a 1-h infusion of acyclovir (5 mg/kg). The 1-h value represents the concentration of drug at the end of the acyclovir infusion. A concentration of 1 μg/ml was equivalent to 4.44 μM.

[b] Mean ± standard error of the mean.

The calculated dose of 5 mg/kg for each patient was withdrawn, and enough Ringers lactate was added to bring the final volume to 60 ml. This was administered by constant intravenous infusion over 1 h (1 ml/min) via a peripheral vein. At 24 to 25 h after the end of the first infusion, each patient received a second infusion administered in an identical fashion, except that the patient was given 1 g of probenecid orally 1 h before the second infusion.

Collection of blood and urine specimens. Venous blood samples for determinations of acyclovir concentrations were drawn through an indwelling catheter (either butterfly INT [Abbott Hospital, Inc., Chicago, Ill.] or Angiocath 20 gauge with a VICRA injection site [Travenol Laboratories, Inc., Deerfield, Ill.]) inserted in the arm contralateral to the infusion site, and samples were placed in 5-ml heparinized tubes. Blood was sampled before infusion, at 0.25, 0.5, and 0.75 h into the infusion, at the end of infusion, and 0.5, 1, 2, 3, 5, 7, 9, 11, 15, and 23 h after the end of infusion. Blood samples were centrifuged, and the plasma was removed and frozen for subsequent determinations of acyclovir concentrations.

Voided urine samples were collected fractionally as follows: a base-line urine sample, 0 to 1 h (during the infusion), 1 to 5 h, 5 to 9 h, 9 to 13 h, and 13 to 25 h after the start of both infusions. Urine samples were also collected 25 to 48 h and 48 to 72 h after the start of the second infusion.

Acyclovir levels. Plasma and urine concentrations were determined by a sensitive and specific radioimmunoassay, as described previously (10). The lower limit of detection of acyclovir by this assay is 0.011 μg/ml (0.05 μM). All samples were assayed in triplicate, and the coefficient of variation was approximately 5%.

Pharmacokinetic analysis. Visual inspection of the plasma concentrations plotted as a function of time on semilogarithmic coordinates indicated that the concentrations appeared to decay in a biexponential manner after the end of the acyclovir infusion period. This pattern is consistent with a two-compartment open model, and therefore the individual patient data were analyzed by a method similar to that described previously (5), using the nonlinear least-squares regression program NONLIN (9) in conjunction with a specific subroutine for the two-compartment open model with zero-order input. A weighting function of 1/concentration was used in the analysis. An exponential curve-stripping program (CSTRIP [12]) provided the initial model parameters for input into the NONLIN program. The best-fit parameter estimates of the elimination rate constant (k_{10}), the intercompartmental transfer rate constants (k_{12}, k_{21}), and the volume of distribution of the central compartment were obtained by NONLIN analysis.

The model-derived parameters, including total body clearance, area under the plasma concentration-time curve from zero to infinity, volume of distribution at steady state, and the beta or terminal half-life, were calculated by standard kinetic equations, as described previously (5, 7). All of the clearance and volume terms were normalized to 1.73 m^2. The body surface area was estimated by the method of Gehan and George (6). Acyclovir renal clearance was determined as previously described (4, 7), based on the amount of acyclovir excreted in the urine in each collection period and the model-fitted calculated area under the curve in the same time period.

Estimates of creatinine clearance were computed based on patient age, weight, and serum creatinine by the method of Mawer et al. (8). When the actual body weight exceeded the calculated lean body weight by 10%, the latter was used to calculate creatinine clearance.

Statistical analysis. The paired Student t test was used to determine the statistical significance of the mean levels and kinetic parameters before and after probenecid treatment. A P value of ≤ 0.05 was considered statistically significant.

RESULTS

The plots of plasma acyclovir concentration versus time for individual subjects showed that probenecid enhanced the plasma concentration of acyclovir after infusion of the drug in all cases. The plasma acyclovir concentrations during and after acyclovir infusion alone and in the presence of probenecid are compared in Table 1. The differences in the mean acyclovir levels between the two groups were not statistically significant at any time up to 2.0. However, at all times after 2.0 h we found statistically significant higher mean acyclovir levels after probenecid administration (Table 1).

The pharmacokinetic data derived from the infusions with and without probenecid are shown in Table 2. In the absence of probenecid, the renal clearance of acyclovir was almost threefold greater than the estimated creatinine clearance, and the total clearance of acyclovir (300 ml/min per 1.73 m^2) was due almost entirely to renal clearance (248 ml/min per 1.73 m^2). The mean urinary recovery of unchanged drug was 79% of the administered dose.

TABLE 2. Pharmacokinetic data for acyclovir in the presence and absence of probenecid[a]

Treatment	$T_{1/2\beta}$ (h)	k_{12} (h^{-1})	k_{21} (h^{-1})	k_{10} (h^{-1})	V_1 (liters/1.73 m^2)	V_{dss} (liters/1.73 m^2)	AUC_0^∞ (μg·h/ml)	Serum creatinine (mg/dl)	Estimated creatinine clearance (ml/min per 1.73 m^2)	Total body clearance (ml/min per 1.73 m^2)	Renal clearance (ml/min per 1.73 m^2)
Acyclovir alone	2.30 ± 0.15[b]	1.16 ± 0.24	0.795 ± 0.117	1.05 ± 0.09	17.1 ± 1.0	41.4 ± 1.0	20.5 ± 2.6	0.73 ± 0.09	90 ± 28	300 ± 40	248 ± 46
Acyclovir + probenecid	2.72 ± 0.18	1.03 ± 0.42	0.807 ± 0.145	0.747 ± 0.177	18.4 ± 3.5	39.2 ± 3.6	28.7 ± 2.7	0.77 ± 0.07	89 ± 29	213 ± 30	168 ± 28

[a] Abbreviations: $T_{1/2\beta}$, terminal half-life; V_1, volume of distribution of the central compartment; V_{dss}, volume of distribution at steady state; AUC_0^∞, area under the plasma concentration-time curve from zero to infinity.
[b] Mean ± standard error of the mean.

After probenecid treatment, the mean terminal plasma half-life of acyclovir increased by 18% (from 2.3 to 2.7 h; $P < 0.01$), and the area under the plasma concentration-time curve from zero to infinity increased by 40% ($P < 0.05$). Probenecid administration had no effect on the apparent volumes of distribution (volume of distribution of the central compartment and volume of distribution at steady state) or any of the rate constants, with the exception of the rate constant of the slow elimination phase. After probenecid administration the mean 25-h acyclovir urinary excretion declined 12.4%, from 79.0 ± 4.9 to 69.2 ± 8.7% of the dose. The total body clearance of acyclovir declined 29% (from 300 to 213 ml/min per 1.73 m^2), and there was a 32% decline in the mean renal clearance of acyclovir after the administration of probenecid. The latter change was statistically significant. However, even after the administration of probenecid, the renal clearance still was about twofold greater than the estimated creatinine clearance.

In none of our patients did we observe any significant adverse reactions during our clinical or laboratory monitoring. We also did not observe any significant extravasation of drug or local intravenous site irritation.

DISCUSSION

Acyclovir is a new potent antiviral drug with potential for widespread use in the treatment of various herpetic infections. Acyclovir elimination occurs predominantly by renal excretion, with about 80% of the dose appearing in the urine as unchanged drug. In a limited number of patients with normal renal function, de Miranda et al. (4) found that 8.5 to 14% of the administered dose appeared as carboxymethoxymethylguanine, the only significant metabolite of acyclovir that has been isolated so far. The finding that the renal clearance of acyclovir is about threefold greater than the creatinine clearance indicates that the renal elimination of acyclovir is only in part by glomerular filtration and that renal tubular secretion plays an important role. In the presence of probenecid, which is a competitive inhibitor of the organic acid secretory system (1), the renal clearance was reduced by about one-third, suggesting that the organic acid transport system is involved in the tubular secretion of acyclovir. However, since the renal clearance of acyclovir was still almost twofold greater than the estimated creatinine clearance, the organic acid transport system may have only a limited role in the elimination of acyclovir. This may be because acyclovir, with pK_a values of 2.27 and 9.25, is both a weak acid and a weak base and may be secreted by other mechanisms. Alternatively, the dose of probenecid given may have been inadequate, and inhibition of the

organic acid transport system may have been incomplete. Perhaps if larger doses of probenecid or repetitive doses of probenecid or both had been administered to our subjects, renal tubular secretion of acyclovir could have been blocked more completely.

Although probenecid does inhibit the elimination of acyclovir, the clinical significance of this effect is probably limited. The increase in the terminal acyclovir plasma half-life is small, and the influence of probenecid on the persistence and clearance of acyclovir, although statistically significant, probably has only limited clinical importance. However, probenecid may allow the persistence of acyclovir levels above some critical value (such as the 50% inhibitory dose for the virus) for a slightly longer period of time, and this may prove to be of limited usefulness.

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