creased gentamicin excretion by a renal tubular process, an event which was not present in the control animals studied. Finally, when water-deprived and hydrated rats were treated with gentamicin (17), the trend in changes seen in Cl_B and Vd with hydration were of similar nature to the animals with glomerulonephropathy. Extrapolation of these findings to the diseased animal is difficult due to the precisely defined experimental conditions of these studies. This is especially true in micropuncture experiments where small quantities of drug are involved. However, these data provide a physiologic basis for the pharmacokinetic changes detected in the dogs and sheep with acute glomerular disease.

It would appear that certain physiologic conditions and disease states associated with volume expansion and/or increased urine flow might lead to increased gentamicin clearance. This response may be modulated through a mechanism triggered by the abnormal fluid status. The increase in drug clearance may be a reflection of nephron heterogeneity (15) or a result of active tubular secretion, decreased proximal tubular reabsorption, or decreased nonionic back diffusion in the distal nephron (16, 18). Alternatively, glomerular disease may specifically increase the drug's ultrafilterability across the glomerular capillary membrane, normally restricted due to the Donnan Effect (18). This functional lesion would be expected to increase the fractional urinary excretion of gentamicin. Mechanistic studies have not been performed in diseased animals, and thus, further speculation is not warranted. This situation may not be seen in the chronic disease situation where individual nephrons have undergone compensatory hypertrophy. This chronic condition is pathophysiologically distinct and is clinically characterized by a different syndrome than is the acute disease process.

In view of these changes, one must be cautious in predicting Cl_B from the glomerular filtration rate, especially when an increased Vd is present, because these methods assume that this relationship does not change with the underlying disease process. However, certain disease conditions may uncouple this association of Cl_B to the glomerular filtration rate. Actual renal clearances of drug should be determined by measuring urinary drug excretion. Note that serum elimination half-life may remain relatively stable in the above situation because the increasing clearance will offset the increased volume of distribution. Dosage nomograms, which correlate elimination half-life or decreasing Cl_B with decreasing creatinine clearance, must be interpreted differently when Vd is known to have increased. Finally, additional studies relating drug disposition to specific pathophysiologic states of renal disease must be conducted to define the effects of various disease processes on drug clearance and volume of distribution, the two physiologic determinants of drug disposition.

(1) J. E. Riviere, G. L. Coppoc, E. J. Hinsman, and W. W. Carlton, *Antimicrob. Agents Chemother.*, **20,** 387 (1981).

(2) S. A. Brown, J. E. Riviere, and G. L. Coppoc, *Proc. Conf. Res. Workers Anim. Dis.*, **62,** 15A (1981).

(3) J. E. Riviere and G. L. Coppoc, *Am. J. Vet. Res.*, **42,** 1621 (1981).

(4) M. Gibaldi and D. Perrier, *J. Clin. Pharmacol.*, **12,** 201 (1972).

(5) V. Klotz, *Clin. Pharmacokinet.*, **1,** 104 (1976).

(6) M. Gibaldi, *Am. J. Med.*, **62,** 471 (1977).

(7) W. J. Jusko and M. Gibaldi, *J. Pharm. Sci.*, **61,** 1270 (1972).

(8) J. C. Pechere and R. Dugal, *Clin. Pharmacokinet.*, **4,** 170 (1979).

(9) P. G. Welling, A. Baumueller, C. C. Lau, and P. O. Nodsen, *Antimicrob. Agents Chemother.*, **12,** 328 (1977).

(10) J. C. Pechere, R. Dugal, and M. M. Pechere, *Clin. Pharmacokinet.*, **3,** 395 (1978).

(11) A. Gyselynck, A. Forrey, and R. Cutler, *J. Infect. Dis.*, **124** (Suppl), S70 (1971).

(12) R. J. Glassock and C. M. Bennett, in "Progress in Glomerulonephritis," P. Kincaid-Smith, A. J. F. d'Apice, and R. C. Atkins, Eds., John Wiley, New York, N.Y., 1979, p. 159.

(13) M. Gibaldi and D. Perrier, "Pharmacokinetics," Marcell Dekker, New York, N.Y., (1975).

(14) E. R. Garrett, *Int. J. Clin. Pharmacol.*, **16,** 155 (1978).

(15) H. O. Senekjian, T. F. Knight, and E. J. Weinman, *Kidney Int.*, **19,** 416 (1981).

(16) C. Carbon, A. Contrepois, A. Vigneron, and S. Lamotte-Barrillon, *J. Pharmacol. Exp. Ther.*, **213,** 600 (1980).

(17) J. LeCompte, L. Dumont, J. Hill, P. Du Souich, and J. Lelorier, *ibid.*, **218,** 231 (1981).

(18) E. Pastoriza-Munoz, D. Timmerman, S. Feldman, and G. J. Kalogomides, *ibid.*, **220** 604 (1982).

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Isosorbide Dinitrate: Pharmacokinetics after Intravenous Administration

Keyphrases □ Pharmacokinetics—intravenous administration of isosorbide dinitrate, bioavailability □ Bioavailability—pharmacokinetics of isosorbide dinitrate after intravenous administration □ Isosorbide dinitrate—pharmacokinetics after intravenous administration, bioavailability

To the Editor:

Isosorbide dinitrate is an organic nitrate found therapeutically useful in its sublingual and oral forms in various cardiovascular diseases such as angina pectoris (1) and congestive heart failure (2). Recently, Distante *et al.* (3) showed that an intravenous infusion of this drug (0.021–0.083 mg/min) was also effective in managing unstable angina. The availability of an intravenous dosage form of isosorbide dinitrate not only affords the opportunity to characterize the pharmacokinetics of this drug after this particular mode of therapy, it also allows the possibility to assess the bioavailability of this drug after other (*e.g.*, oral) routes of administration in patients. This latter subject has been of major controversy since Needleman *et al.* (4) made the assertion that oral nitrate therapy is irrational because of its complete first-pass metabolism.

A preliminary study has appeared which provided some initial information on this important issue. Taylor *et al.* (5) showed that in two normal, young subjects who received

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intravenous isosorbide dinitrate, the plasma clearance was found to be 0.32 and 0.16 liter/min. Based on these values, these authors concluded that sublingual and oral administration of isosorbide dinitrate are only bioavailable to the extent of 6 and 3%, respectively. If these data can be confirmed in a larger group, they could represent an unusual example of very poor sublingual bioavailability of a small neutral compound given at a relatively low dose. Further, if shown applicable to patients, these data could also have wide-ranging implications regarding the clinical use as well as the regulatory policy of oral and sublingual dosage forms of isosorbide dinitrate. It follows then that there may be a necessity (and perhaps urgency) of further research in identifying the reasons for, and approaches in circumventing, the poor sublingual bioavailability of this drug.

We have expanded on the study of Taylor *et al.* (5) through an examination of the pharmacokinetics of isosorbide dinitrate in 11 cardiac patients after intravenous infusion at two different rates. These data were obtained as part of a comprehensive study which examined the pharmacokinetics of isosorbide dinitrate after intravenous, sublingual, oral, and percutaneous administration. Because the intravenous data are crucial in the strategic planning of further pharmaceutical research of this drug, we present here a preliminary report on this aspect for the interest of workers in this area.

Eleven angina patients were infused intravenously with an isosorbide dinitrate solution[1] which had been suitably diluted such that a total dose of 2 mg in 10 ml was delivered over 15 min (infusion rate = 0.133 mg/min). Two of these patients received isosorbide dinitrate again on another day at a slower infusion rate (0.083 mg/min), one for 1 hr and the other for a 2-hr duration. This latter infusion rate was identical to that used by Taylor *et al.* (5). The strength of the dosing solution, *via* an HPLC analysis developed in this laboratory[2], was determined to be at a mean of 94% of the theoretical value. The concentration of isosorbide dinitrate in the infusion solution emerging from the polyethylene infusion tubing was essentially identical both before and after the experiment, indicting that adsorptive loss to the infusion system was absent. Serial plasma samples were drawn, anticoagulated with sodium citrate, and frozen (−20°) until assayed for isosorbide dinitrate by GC–electron capture detection (6).

The terminal disapearance phase rate constant (β) was obtained by least-square regression of the last four concentration–time points. The area under the plasma concentration *versus* time curve from time zero to the last plasma sample collected (C_t) was determined by Lagrange integration (7). Total area under the curve (AUC) was estimated by addition of the residual area (~15%) which was calculated by dividing C_t by β. The systemic clearance (Cl_{iv}) was calculated by dividing the administered dose by AUC. The apparent volume of distribution (Vd_{area}) was then determined by dividing the systemic clearance by β (8).

Figure 1 shows the plasma concentration of isosorbide dinitrate after intravenous infusion in the patient population studied. Results from the short infusion (2 mg over 15 min) showed that steady-state concentrations were not

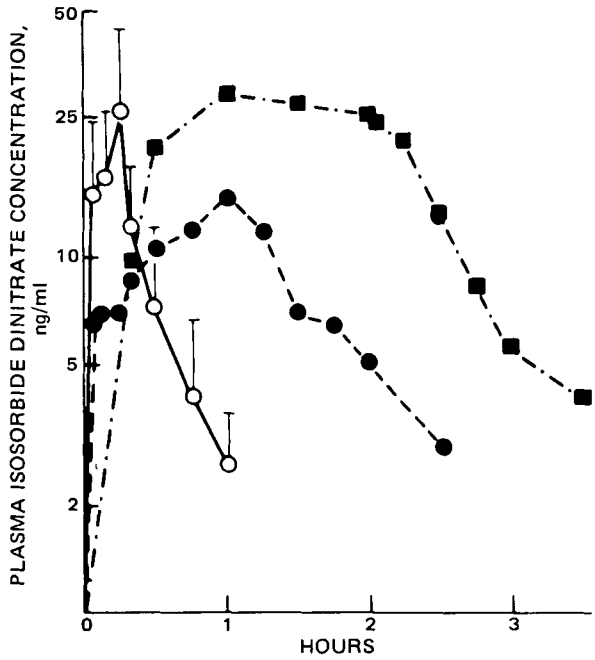

Figure 1—*Plasma isosorbide dinitrate concentrations observed after different rates and duration of intravenous infusion. Key: (○) 0.133 mg/min for 15 min, mean +SD, n = 11, (●) 0.083 mg/min for 1 hr, (■) 0.083 mg/min for 2 hr.*

achieved after this infusion regimen and that decline of plasma drug was apparently biexponential. Unfortunately, the initial rapid distributive phase observed after cessation of the infusion could not be quantitated due to insufficient sampling. The mean half-life of the terminal disappearance phase, however, was found to be 18 ± 7 min (± SD, n = 11), with a range of 10–30 min. The Cl_{iv} value was estimated at 3.4 ± 1.4 liter/min (mean ± SD, n = 11) with a range of 1.8–6.3 liter/min. The mean Vd_{area} (±SD) was estimated to be 101 ± 67 liters (range 46–241 liters). Infusion rate and duration did not appear to affect the pharmacokinetics of isosorbide dinitrate: in one patient, Cl_{iv} was estimated at 4.0 and 4.7 liter/min at infusion rates of 0.083 mg/min for 1 hr and 0.133 mg/min for 15 min, respectively; in a second patient, Cl_{iv} was 2.9 and 2.7 liter/min at infusion rates of 0.083 mg/min for 2 hr and 0.133 mg/min for 15 min, respectively.

If a prolonged elimination phase existed, but was undetected due to assay limitations, the Cl_{iv} values obtained here would be overestimated. However, in the patient infused for 2 hr at 0.083 mg/min, both the time to steady state, and concentration at steady state were consistent with the experimentally observed β and Cl_{iv} values, respectively. Regardless, a doubling of the terminal half-life for residual area calculation would only lead to a 7% decrease in Cl_{iv}. These observations suggest that the Cl_{iv} values obtained here are both internally consistent and reliable.

The clearance values observed in our patient population were, in general, at least 10-fold larger than those obtained by Taylor *et al.* (5) in two normal subjects. The reasons for this discordance are not known; however, a study is in progress which will reassess the validity of the low bioavailability estimates of sublingually and orally administered isosorbide dinitrate reported thus far, particularly to its applicability in cardiac patients.

[1] Isoket Ampullen, Pharma-Schwarz GmbH, 4019 Monheim, West Germany.

[2] H.-L. Fung and S. L. Chan, unpublished observation.

(1) T. Sweatman, G. Strauss, A. Selzer, and K. E. Cohn, *Am. J. Cardiol.*, **29**, 475 (1972).
(2) J. A. Franciosa, E. Mikulic, J. N. Cohn, E. Jose, and A. Fabie, *Circulation*, **50**, 1020 (1974).
(3) A. Distante, A. Maseri, S. Severi, A. Biagini, and S. Chierchia, *Am. J. Cardiol.*, **44**, 533 (1979).
(4) P. Needleman, S. Lang, and E. M. Johnson, *J. Pharmacol. Exp. Ther.*, **181**, 489 (1972).
(5) T. Taylor, L. F. Chasseaud, and E. Doyle, *Biopharm. Drug Dispos.*, **1**, 149 (1980).
(6) R. A. Morrison and H.-L. Fung, in "Abstracts", Vol. 10, No. 1, APhA Academy of Pharmaceutical Sciences, Washington, D.C., 1980, p. 126.
(7) K. C. Yeh and K. C. Kwan, *J. Pharmacokinet. Biopharm.*, **6**, 79 (1978).
(8) M. Gibaldi and D. Perrier, Eds., "Pharmacokinetics," Marcel Dekker, New York, N.Y., 1975.

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Time-Dependent Kinetics VIII: Absence of Diurnal Oscillations in Valproic Acid Disposition Following Single Dose Administration to Rhesus Monkey

Keyphrases □ Valproic acid—time-dependent kinetics, absence of diurnal oscillations, disposition following single dose administration to rhesus monkey □ Kinetics, time-dependent—absence of diurnal oscillations in valproic acid disposition following single dose administration to rhesus monkey

To the Editor:

Valproic acid exhibits an unusual pharmacokinetic property in the rhesus monkey, namely, extensive diurnal oscillations in systematic clearance. An initial study in three normal rhesus monkeys showed that during constant rate intravenous infusion, steady-state levels increased at night with maxima 40–140% higher than the corresponding minima (1). In a subsequent study in four normal monkeys, where levels were monitored for 48 hr, it was found that the diurnal fluctuations in steady-state levels were reproducible in 2 consecutive days (2). Furthermore, after reversal of the 12-hr light–12-hr dark cycle, plasma concentrations tended to follow the phase shift with maxima during the reversed dark phase (actual day time) (2). In a later efficacy study in 12 epileptic monkeys, valproic acid was infused at a constat rate to achieve steady-state levels of 46, 97, and 147 μg/ml (3). The diurnal fluctuations in valproate levels were found at all three steady-state concentrations (4).

In each of the studies just described, the diurnal changes in valproate clearance were found under steady-state conditions. In the present study, the objective was to determine whether this phenomenon could be observed with an acute mode of administration. To this effect, valproate was administered at different times of day by intravenous boluses to a group of six rhesus monkeys. In addition, this design would allow a detection of diurnal effects in valproate distribution.

Six chair-adapted male rhesus monkeys (mean body weight 4.1 kg) with two chronic venous catheters (femoral for valproate bolus injection and jugular for blood sampling) were used in this study. Environmental conditions were maintained the same as those described previously (1, 2) (diurnal cycle: light period, 6 am–6 pm; dark period, 6 pm–6 am). Based on the findings of previous studies (valproate plasma levels remained stable or decreased during 10 am–6 pm, increased and reached a maximum during 6 pm–6 am, and tended to decline from 6 am–noon), the following times were selected for drug administration: 2 am, 8 am, 2 pm, and 8 pm. At these times of the day, valproate intravenous bolus injections were administered in a randomized fashion to six monkeys. At least 1 week of rest was allowed between any two injection times. From 2 to 4 replicate studies at each time period were conducted for each of the six monkeys.

At each time period, including replications, each monkey received 63.75 mg of valproic acid equivalent as sodium salt in 0.5 ml of sterile saline as an intravenous bolus injection. An additional 5 ml of saline was used to flush the line after bolus administration. Blood samples (2 ml) were collected in vacuum tubes containing edetic acid at 2, 20, 40, 60, and 100 min following drug administration. Plasma was separated and frozen until assay. Valproate was assayed by GLC using the procedure of Levy *et al.* (5).

The area under the plasma concentration–time curve (extrapolated to infinite time) was calculated by the trapezoidal rule and the systemic or total body clearance was computed from the dose–area relationship. The plasma valproate concentration time data were least-squares fitted to a monoexponential decay equation (BMDX-85), and the volume of distribution was calculated by the ratio of total body clearance and elimination rate constant. The null hypothesis of equal mean values for clearance, volume of distribution, and elimination rate constant among treatments was tested using a one-way ANOVA for repeated measures (BMDP2V). Then Tukey's method for multiple comparisons (6) was used to test for differences between particular pairs of treatments.

Mean plasma valproate concentration–time profiles at 2 am, 8 am, 2 pm, and 8 pm are shown in Fig. 1. Based on the findings of previous studies, which revealed the presence of circadian rhythms in steady-state valproate plasma levels (1–4), lower clearance values would be expected at 2 am and 8 pm than at 2 pm and 8 am. However, no significant difference in valproate clearance was observed between any of the four time periods. Volume of distribution and half-life also did not exhibit any time dependence ($p > 0.05$). Values of clearance, volume of distribu-

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