Br. J. clin. Pharmac. (1981), **12**, 841–844

PHARMACOKINETICS OF DIAZEPAM IN EPILEPTIC PATIENTS AND NORMAL VOLUNTEERS FOLLOWING INTRAVENOUS ADMINISTRATION

SORAYA DHILLON & ALAN RICHENS

Department of Pharmacology and Materia Medica, Welsh National School of Medicine, Heath Park, Cardiff CF4 4XN

1 The pharmacokinetics of diazepam following intravenous administration have been investigated in six normal volunteers and nine epileptic patients receiving chronic antiepileptic drug therapy.

2 After intravenous administration, serum diazepam levels declined biexponentially in all subjects. The elimination half-life was significantly shorter and the plasma clearance significantly higher in the patients than in the normal volunteers.

3 Serum *N*-desmethyldiazepam levels were higher and the time to peak serum concentration was earlier in the epileptic patients than in the controls.

4 It is suggested that the metabolism of diazepam is induced in patients treated with enzyme inducing antiepileptic drugs, although a protein binding interaction between valproic acid and diazepam may contribute to the higher plasma clearance in the epileptic patients taking sodium valproate.

Introduction

Intravenous diazepam is recognised as a drug of choice for the treatment of status epilepticus although chronic oral administration has proved poorly effective in the control of major seizures. Diazepam is extensively metabolised by demethylation and hydroxylation to the major metabolites, *N*-desmethyldiazepam and 3-hydroxydiazepam. These are then further transformed to oxazepam, which is conjugated with glucuronic acid and excreted in the urine. Therefore changes in drug metabolism caused by hepatic disease or enzyme induction are likely to alter the elimination of diazepam. Studies in animals with enzyme inducing agents have shown stimulation of diazepam metabolism affecting different pathways. Following chronic phenobarbitone administration both *N*-desmethylation and hydroxylation to oxazepam were increased in rats. However, in mice and guinea pigs only one metabolic pathway, either *N*-desmethylation or hydroxylation was influenced by phenobarbitone administration (Marucci *et al.*, 1970). A recent study in humans has shown that the elimination of diazepam and *N*-desmethyldiazepam is significantly increased by induction of liver microsomial enzymes following administration of antipyrine as the inducing agent (Ohnhaus *et al.*, 1979). However the effect of enzyme inducing antiepileptic drugs on diazepam metabolism in man has not been investigated. We have therefore studied the disposition of diazepam in a group of epileptic patients receiving concurrent antiepileptic drug therapy and compared this with the dispositon of diazepam in healthy volunteer subjects.

Methods

Six healthy drug-free male volunteers, and nine male epileptic patients resident at the Chalfont Centre for Epilepsy, Chalfont St Peter, Bucks., gave their informed consent to take part in the study. Age and body weight were similar in the two groups (Tables 2 and 3). The patients were receiving regular antiepileptic drug treatment (Table 1).

The subjects received a light breakfast of toast and fruit juice in the morning of the test day. Each received 10 mg of diazepam (Valium, Roche) by intravenous injection over a 2 min period. The subjects remained supine during the next 3 h and no fluid or food was permitted during this time. Blood samples (5 ml) were taken from the epileptic patients and normal volunteer subjects at frequent intervals for up to 48 and 96 h respectively. Blood samples on the first day were taken from the arm not used for intravenous injection, using an indwelling butterfly needle.

Serum diazepam and *N*-desmethyldiazepam concentrations were measured by gas chromatography.

0306–5251/81/120841–04 $01.00

Table 1 Details of the drug treatment of the epileptic patients.

Patient	*Regular anticonvulsant treatment (mg total daily dose)*		
1	Carbamazepine	600, sodium valproate	1000
2	Carbamazepine	1200, sodium valproate	1000
3	Sodium valproate	2000	
4	Carbamazepine	800, phenytoin	400
5	Carbamazepine	1000, sodium valproate	2000
6	Carbamazepine	1000, clonazepam	4
7	Carbamazepine	1200, sodium valproate	500
8	Carbamazepine	1200, sodium valproate	1500
9	Primidone	750, sodium valproate	500

The gas chromatograph was a Perkin Elmer F33 fitted with an electron capture detector. A coiled glass column 3 m × 3 mm i.d. was packed with Kovats 3%C_{87} on Chromosorb W 100-120 mesh. The temperature of the column was 255°C and the temperature of the injection port and detector was 300°C. Nitrogen gas flow was 120 ml min^{-1}. The extraction procedure followed was as described by Rutherford (1977). Within assay variability between batches was found to be 4.4% and 2.2% (coefficient of variation) for serum samples containing 25 ng ml^{-1} and 500 ng ml^{-1} respectively.

Analysis of data

Following intravenous administration the serum diazepam concentration declined biexponentially in all subjects. Data were analysed according to a two-compartment open model. According to this model, at time t after a single intravenous dose, the serum concentration C_t is given by the equation

$$C_t = A.e^{-\alpha t} + B.e^{-\beta t}$$

where α and β are the rate constants of the initial rapid and terminal slower phase respectively and A and B the contribution of the corresponding exponentials at t = 0. The β slope was obtained by linear regression from the terminal part of the serum concentration curve and extrapolated back to zero time. The α slope was calculated by linear regression from the values obtained after subtracting β slope to the serum concentration of the early samples.

The area under the serum concentration-time curve was estimated by the trapezoidal rule, and extrapolated to infinity using the equation

$$AUC_{t\to\infty} = \frac{C_t}{\beta}$$

C_t = serum concentration at the last sampling time. Volume of distribution ($V_d\beta$) was calculated from the formula

$$V_d = \frac{\text{Dose}}{AUC_{0\to\infty}\,\beta}$$

and total body clearance (Cl) was calculated according to the model independent formula

$$Cl = \frac{\text{Dose}}{AUC_{0\to\infty}}$$

(Gibaldi, Nagashima & Levy, 1969). Statistical analysis was evaluated using Student's *t*-test.

Results

Mean serum diazepam concentrations following intravenous administration in healthy subjects and epileptic patients are shown in Figure 1. All subjects show a distribution phase virtually complete after 6 h, following which the decline of the log serum concentration-time profile appeared linear. Pharmacokinetic parameters calculated for the two groups of subjects are shown in Tables 2 and 3. In all subjects the serum diazepam concentrations declined biexponentially; in two epileptic subjects however slight irregularities in the early part of the serum concentration curve prevented an accurate estimation of the α phase.

The elimination half-life diazepam was significantly shorter and the total body clearance (Cl) significantly higher in the epileptic patients compared

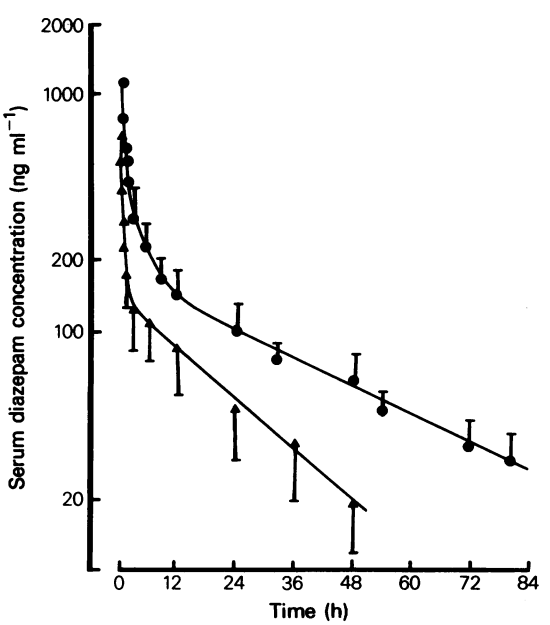

Figure 1 Mean (± s.d.) serum diazepam concentrations in nine epileptic patients (▲) and six healthy volunteers (●) following intravenous administration of diazepam (10 mg).

Table 2 Kinetic parameters calculated from serum diazepam concentration after intravenous administration of 10 mg in epileptic patients.

Patient	Age (years)	Weight (kg)	A (ng ml^{-1})	α (h^{-1})	$T_{½}\alpha$ (h)	B (ng ml^{-1})	β (h^{-1})	$T_{½}\beta$ (h)	$V_d\beta$ (l kg^{-1})	Cl (l h^{-1})
1	19	54	603	7.92	0.09	179	0.05160	13.4	0.90	2.54
2	20	66	—	—	—	179	0.05161	13.4	1.10	3.52
3	22	65	1053	3.84	0.18	123	0.03150	22.0	1.17	2.40
4	25	64	—	—	—	225	0.06030	11.5	0.56	2.15
5	30	76	1177	6.6	0.11	78	0.04751	14.6	1.36	4.83
6	33	67	841	12.60	0.06	311	0.04551	15.2	0.60	1.68
7	35	64	844	2.55	0.27	148	0.05380	12.9	0.96	3.30
8	37	74	909	3.90	0.18	89	0.08310	8.4	0.56	3.46
9	39	67	661	3.61	0.19	231	0.10030	6.9	0.61	4.09
Mean	28	66	870*	5.86*	0.15	174	0.05830	13.1*	0.86	3.10*
s.d.	7.6	6.3	202	3.52	0.07	74	0.02090	4.3	0.29	1.0

*$P < 0.01$ compared with data in volunteers (Table 3)

to the normal healthy subjects. The apparent volume of distribution was similar for the two groups. However, if all the subjects are divided into two groups according to whether they were receiving sodium valproate or not, the former group showed a trend towards higher volumes of distribution (0.95 ± 0.28 and 0.75 ± 0.29 l kg^{-1} respectively).

The mean serum concentration time profile of the major metabolite *N*-desmethyldiazepam in healthy volunteers and epileptic subjects is shown in Figure 2. Peak serum *N*-desmethyldiazepam concentrations were attained earlier in the epileptic subjects and the serum levels were higher at each sampling point up to 48 h when compared with the normal volunteers ($P < 0.05$).

Discussion

This study has shown that the elimination half-life of diazepam in epileptic patients is significantly shorter than in drug free volunteers and the plasma clearance is increased about three fold. Since all but one epileptic subject was receiving an antiepileptic drug which is known to induce hepatic microsomal enzymes (Perucca *et al.*, 1979) it is likely that enzyme induction is the mechanism responsible. Sodium valproate has been shown not to be an enzyme inducing agent (Oxley *et al.*, 1979) and the only patient receiving this drug alone had the largest elimination half-life and one of the lowest clearances of the epileptic subjects. The mean pharmacokinetic parameters that we have found in our normal volunteers are similar to values reported previously (Greenblatt *et al.*, 1980; Mandelli, Tognoni & Garattini, 1980).

That induction of diazepam metabolism is the likely explanation for these findings is supported by the observation that serum *N*-desmethyldiazepam levels were higher and peak serum concentrations were reached earlier in the epileptic patients than in the volunteer subjects, with the exception of Patient 3, the patient not receiving enzyme inducing therapy. Animal experiments have shown that phenobarbitone administration increases *N*-desmethyldiazepam and oxazepam formation in rats (Marucci *et al.*, 1970). In this study we have not attempted to measure oxazepam, but it would be of interest to investigate whether the urinary output of this metabolite, which is the hydroxy derivative of *N*-desmethyldiazepam, is increased in enzyme induced subjects. Furthermore,

Table 3 Kinetic parameters calculated from serum diazepam concentration after single intravenous administration 10 mg in healthy volunteers.

Patient	Age (years)	Weight (kg)	A (ng ml^{-1})	α (h^{-1})	$T_{½}\alpha$ (h)	B (ng ml^{-1})	β (h^{-1})	$T_{½}\beta$ (h)	$V_d\beta$ (l kg^{-1})	Cl (l h^{-1})
1	21	70	1264	3.98	0.17	260	0.03864	17.9	0.45	1.21
2	22	68	1095	3.17	0.22	318	0.02684	25.8	0.43	0.78
3	26	64	1052	1.51	0.46	150	0.02531	27.4	0.84	1.36
4	26	75	921	3.39	0.20	105	0.01586	43.7	1.29	1.54
5	30	73	1737	3.65	0.19	131	0.01227	56.4	0.86	0.77
6	35	75	1181	3.48	0.20	132	0.02102	33.0	0.96	1.53
Mean	27	71	1207	3.20	0.24	183	0.02332	34.0	0.80	1.20
s.d.	5	4	282	0.87	0.11	86	0.00931	12	0.33	0.35

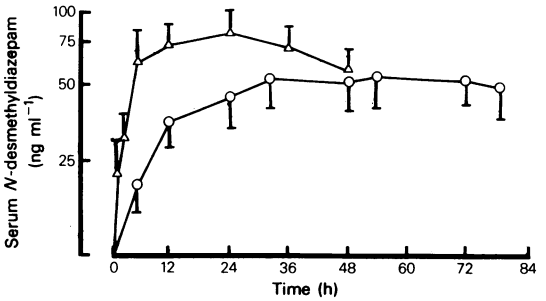

Figure 2 Mean (± s.d.) serum *N*-desmethyldiazepam concentrations in nine epileptic patients (Δ) and six healthy volunteers (O) following intravenous administration of diazepam (10 mg).

there is no evidence available on the possible induction of the 3-hydroxy pathway of diazepam to temazepam.

In our epileptic patients taking sodium valproate, there was a trend towards a higher apparent volume of distribution in comparison to subjects not taking this drug. Valproic acid is a two chain fatty acid with similar properties to endogenous free fatty acids (Monks & Richens, 1979) and has been shown to displace diazepam from protein binding sites (Sjöholm *et al.*, 1979; Dhillon & Richens, 1981). Although this protein binding interaction might be expected to contribute to the higher plasma clearance in the epileptic patients, it would not account for the earlier and higher peak serum *N*-desmethyldiazepam levels seen in these subjects. Enzyme induction by the antiepileptic drugs, therefore remains as the most plausible explanation of the charges observed.

We thank the National Fund for Research into Crippling Diseases for financial support and Dr Jolyon Oxley, Senior Physician at the Chalfont Centre for Epilepsy for allowing us to study patients under his care.

References

COLBURN, W.A. & GIBALDI, M. (1978). Plasma protein binding of diazepam after a single dose of sodium oleate. *J. pharm. Sci.*, **67**, 891–892.

DHILLON, S. & RICHENS, A. (1981). Serum protein binding of diazepam and its displacement by valproic acid *in vitro*. *Br. J. clin. Pharmac.*, **12**, 591–592.

GIBALDI, M., NAGASHIMA, R. & LEVY, G. (1969). Relationship between drug concentration in plasma or serum and amount of drug in the body. *J. pharm. Sci.*, **58**, 193–197.

GREENBLATT, D.J., ALLEN, M.D., HARMATZ, J.S. & SLADER, R.I. (1980). Diazepam disposition determinants. *Clin. Pharmac. Ther.*, **27**, 301–312.

MANDELLI, M., TOGNONI, G. & GARATTINI, S. (1978). Clinical pharmacokinetics of diazepam. *Clin. Pharmacokin.*, **3**, 72–91.

MARUCCI, F., FANELLI, R., MUSSINI, E. & GARATTINI, S. (1970). Effect of phenobarbital on the *in vivo* metabolism of diazepam in several animal species. *Biochem. Pharmac.*, **19**, 1771–1776.

MONKS, A. & RICHENS, A. (1979). Serum protein binding of valproic acid and its displacement by palmitic acid *in vitro*. *Br. J. clin. Pharmac.*, **8**, 187–189.

OHNHAUS, E.E., PARK, B.K., COLOMBO, S.P. & HEIZMANN, P. (1979). The effect of enzyme induction on diazepam metabolism in man. *Br. J. clin. Pharmac.*, **8**, 557–563.

OXLEY, J., HEDGES, A., MAKKI, K., MONKS, A. & RICHENS, A. (1979). Lack of hepatic enzyme inducing effect of sodium valproate. *Br. J. clin. Pharmac.*, **8**, 189–190.

PERUCCA, E., HEDGES, A., MAKKI, K., HEBDIGE, S., WADSWORTH, J. & RICHENS, A. (1979). The comparative enzyme-inducing properties of antiepileptic drugs. *Br. J. clin. Pharmac.*, **7**, 414P–415P.

RICHENS, A. (1979). The comparative enzyme inducing properties of antiepileptic drugs. *Br. J. clin. Pharmac.*, **7**, 414P–415P.

RUTHERFORD, D.M. (1977). A rapid micro-method for the measurement of diazepam and *N*-desmethyldiazepam in blood, plasma by gas liquid chromatography. *J. Chromat.*, **137**, 439–448.

SJÖHOLM, I., EDMAN, B., KOBER, A., LJUNGSTEDT-PAHLMAN, I., SEIVING, B. & SJÖDIN, T. (1979). Binding of drugs to human serum albumin: XI The specificity of three binding sites as studied with albumin immobilised in micropeptides. *Mol. Pharmac.*, **16**, 767–777.

(Received May 14, 1981)