PLASMA CONCENTRATIONS OF DIAZEPAM AND DESMETHYLDIAZEPAM DURING CHRONIC DIAZEPAM THERAPY

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- 1 Plasma concentrations of diazepam and its metabolite, desmethyldiazepam, have been measured in both in- and out-patient groups treated with diazepam for periods varying between 1 month and 10 years.
- 2 The diazepam concentration was directly related to the dose of diazepam ingested and inversely related to the age of the patient.
- 3 A highly significant relationship was obtained between the concentrations of desmethyldiazepam and diazepam.
- 4 The plasma concentrations of both diazepam and its metabolite were independent of sex, duration of therapy and patient group.

Introduction

Diazepam is used widely in the treatment of insomnia and anxiety. For some patients, this treatment is prolonged, often continuing for several years. Although the pharmacokinetics of diazepam and its metabolites have been studied extensively following both single and multiple doses of the drug (Kaplan, Jack, Alexander & Weinfeld, 1973; Hillestad, Hansen, Melsom and Drivenes, 1974; Hillestad, Hansen & Melsom, 1974; Klotz, Avant, Hoyumpa, Schenker & Wilkinson, 1975; Klotz, Antonin & Bieck, 1976), few multiple dose studies have been extended beyond two weeks.

In one study in which patients had received diazepam therapy of from 1 to 6 weeks duration, a decrease in the steady-state plasma concentrations of both diazepam and desmethyldiazepam was observed (Kanto, Iisalo, Lehtinen & Salminen, 1974). These workers also found that the plasma concentrations of both drug and major metabolite were higher in patients who had received diazepam therapy for only two weeks, compared to those who had been treated continuously with diazepam for several months or years. However, Zingales (1973) was unable to find a consistent relationship between the plasma concentration of diazepam and the duration of therapy. In this

latter study, marked individual differences in the plasma concentrations of both diazepam and desmethyldiazepam were observed in patients treated with the same dose of drug. Despite these variations, the average plasma concentrations appeared to be related to the dose received.

Although it has been shown that the plasma concentrations of diazepam are not directly related to the drug's clinical effects after 2–4 weeks therapy (Bond, Hailey & Lader, 1977), the measurement of such concentrations may still be of assistance to the clinician, especially in the assessment of patient compliance and drug abuse. However, to use the analytical results for this purpose it is necessary to know how the plasma concentration of diazepam is related to dosage. Thus, the initial aims of the present study were to investigate this relationship and also, to ascertain whether any other variables (especially duration of therapy) were significant predictors of the plasma concentration.

Methods

Two groups of patients (I and O) were studied. Group I consisted of twenty in-patients at a psychiatric hospital where they had been for at least 6 weeks. Each patient had been treated with diazepam continuously during this period and confirmation that their prescribed dose had been ingested was obtained from the nurses' records. Any other medication ingested by these patients was recorded also. Group O

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consisted of sixteen out-patients who were attending a psychiatric clinic. Most of the patients were prescribed diazepam alone and all had been receiving this drug for a minimum of 6 weeks. Only those patients who admitted to having taken their medication regularly were included in the study. However, no independent check on drug ingestion was available. The main demographic features of both groups are shown in Table 1.

After the withdrawal of a venous blood sample (5 ml) from each patient, the plasma was separated immediately and then frozen until sent for analysis. From group I patients, blood samples were obtained in the morning at least ten hours after their latest diazepam dose, whereas from group O patients, samples were collected at different times during the day and thus, with the latter group, there was no consistent interval between the time of the patient's latest dose and that of venepuncture. Plasma concentrations of diazepam and its metabolite, desmethyldiazepam were measured using a gas-liquid chromatographic procedure (Rutherford, 1977).

For each patient the following variables were recorded: sex, age, daily diazepam dose, duration of continuous diazepam therapy, plasma concentrations of both diazepam and desmethyldiazepam, and group. Multiple regression analyses of the data were performed using the BMD Programs, P1R and P2R.

Results

The results obtained for each patient are presented in Table 2. Multiple regression analysis of the combined data was used to ascertain which of these variables were significant predictors of the plasma concentration of diazepam. Before the data was analyzed in this way, the distribution of the dependent variable was examined and found to be significantly different at the 1% level (one-tailed test) from the value expected for a

normal distribution. However, square root transformation of this variable was effective in stabilizing the variance, and thus the transformed values were used in subsequent calculations.

The regression equation that gives the best prediction of the values of the plasma diazepam concentration (D), when the plasma concentrations of desmethyldiazepam were excluded from the analysis, was:

$$D^{0.5} = 15.066 + 0.700 (\text{dose}) - 0.149 (\text{age})$$

Dose was found to be the primary variable in this equation irrespective of the order in which the variables were entered in the multiple regression calculation. The variance ratio (VR) was highly significant (VR=50.50, $P < 0.001$) and the squared multiple correlation coefficient ($r^2 = 0.75$) indicated that the predictive value of the variables, dose and age, was quite high. Both partial regression coefficients were significantly different from zero ($t = 8.84$, $P < 0.001$ and $t = 3.56$, $P = 0.001$ for dose and age respectively) and thus, each predictor variable contributed independently to the effectiveness of the overall regression.

The plasma concentration of desmethyldiazepam (DD) correlated with D ($r = 0.87$, $P < 0.001$) and could be predicted from the equation:

$$(DD)^{0.5} = 12.289 + 0.031 (D)$$

The distribution of the values of DD was positively skewed, however square root transformation was effective in stabilizing the variance. For this regression analysis only the values of the dependent variable were transformed. The variance ratio was highly significant (VR=105.67, $P < 0.001$) and 76% of the variation in $(DD)^{0.5}$ was explained by the inclusion of D in the regression equation. The regression coefficient was significantly different from zero ($t = 10.28$, $P < 0.001$). None of the other variables were found to significantly affect the prediction of DD.

Table 1 Summary of age, sex, daily diazepam dose and duration of therapy for both groups of patients included in this survey

Variable	Units	In-patients (n=20)	Out-patients (n=16)	All subjects
Age (mean $\pm$ s.d.)	years	57.3 $\pm$ 11.9	40.9 $\pm$ 10.6	50.0 $\pm$ 13.9
Range	years	29-72	24-60	24-72
Sex				
Male		10	4	14
Female		10	12	22
Daily diazepam dose (mean $\pm$ s.d.)	mg	11.9 $\pm$ 6.1	15.3 $\pm$ 8.6	13.4 $\pm$ 7.4
Range	mg	4-30	2-30	2-30
Duration of therapy (mean $\pm$ s.d.)	months	24.0 $\pm$ 27.8	43.9 $\pm$ 38.3	32.8 $\pm$ 33.9
Range	months	1-84	2-120	1-120

The above regression equations were calculated on the assumption that there was no significant difference between the plasma concentrations measured in the two groups, despite the variations in the times of blood sampling. The validity of this assumption was

confirmed by comparing the regression of (DD) on $D^{0.5}$ in each group; for group I, $(DD)^{0.5} = 12.007 + 0.030 D$ and group O, $(DD)^{0.5} = 13.679 + 0.030 D$. Since neither the regression coefficients nor the intercepts were

Table 2 Plasma concentrations of diazepam (D) and desmethyldiazepam (DD) measured in patients on chronic diazepam therapy

Number	Daily dose (mg)	Age (years)	Group (I/O)*	Sex	Duration of therapy (months)	D ($\mu\text{g/l}$)	DD ($\mu\text{g/l}$)	Other drugs
1	2	24	O	F	3	138	274	—
2	4	71	I	F	23	42	95	phenobarbitone
3	5	62	I	M	8	95	156	amitriptyline
4	5	57	I	M	60	66	149	—
5	6	60	I	F	5	123	286	amitriptyline potassium citrate
6	6	46	O	F	9	214	692	—
7	7	69	I	M	6	20	55	phenytoin clofibrate
8	8	29	I	F	6	175	225	orphenadrine nitrazepam flupenthixol
9	8	35	O	M	24	236	494	—
10	8	29	O	M	48	234	365	—
11	10	64	I	M	2	212	268	amitriptyline
12	10	53	I	M	4	100	255	phenytoin flupenthixol
13	10	50	O	F	9	225	277	—
14	10	53	I	F	40	49	109	chlorpromazine tetrabenazine penicillin
15	10	65	I	M	72	185	360	amitriptyline
16	10	48	O	F	84	179	113	—
17	11	63	I	F	62	172	417	—
18	12.5	72	I	M	1	195	229	—
19	15	51	I	M	1	424	1360	amitriptyline disulfiram
20	15	28	O	F	2	502	727	—
21	15	56	I	F	2	504	551	—
22	15	62	I	F	5	389	804	amitriptyline
23	15	52	I	M	7	379	448	ethinyloestradiol penicillin
24	15	60	O	F	24	229	586	—
25	15	35	O	F	37	559	852	—
26	15	48	O	F	48	669	1090	imipramine
27	15	42	I	M	60	310	512	—
28	15	61	I	F	84	350	807	amitriptyline
29	15	40	O	M	96	424	872	—
30	15	27	O	F	96	704	951	—
31	20	71	I	F	13	255	541	iprindole flurazepam
32	20	43	O	F	120	463	977	imipramine
33	30	46	O	F	12	737	1146	phenelzine
34	30	40	O	F	18	454	757	—
35	30	32	I	F	18	945	1040	cortisone thioridazine fludrocortisone
36	30	56	O	M	72	1010	1765	—

* I = in-patient; O = out-patient

significantly different ($VR=0.6$, $P=0.5$), it is likely that the majority of the plasma concentrations measured in both groups were close to their steady-state values.

Discussion

Multiple regression analysis of the data revealed the existence of a significant relationship between the daily dose of diazepam and the plasma diazepam concentration; this result confirms the observations of Zingales (1973). Thus, the measurement of plasma diazepam concentrations can provide a means of assessing patient compliance and may be especially helpful in cases of suspected diazepam abuse. The fact that the duration of therapy had no significant effect on the above relationship conflicts with the results obtained by Kanto *et al.* (1974). At equivalent dosages, the plasma concentrations measured by the latter authors were much lower, on average, than those obtained both in our study and by Zingales (1973). Although the number of patients studied by Kanto *et al.* (1974) was small, poor compliance of patients may have accounted for these differences in concentrations.

The association of decreasing plasma concentrations with increasing age during chronic diazepam therapy has not been reported previously for this drug. Lower plasma concentrations of the drug have been found in elderly patients after an acute dose of diazepam (Garattini, Marcucci, Morselli & Mussini, 1973; Klotz *et al.*, 1975); this has been attributed to age-dependent changes in the volume of distribution of diazepam. However, since plasma clearance was independent of age, these latter authors predicted that the steady-state concentrations achieved during chronic diazepam therapy would also be independent of age. Whether these concentrations can be accurately predicted from single dose studies, considering the complex nature of the pharmacokinetics of diazepam, must be doubtful,

especially since a recent investigation (Klotz *et al.*, 1976) showed that, after sub-chronic doses of diazepam, the plasma clearance of the drug decreases, thereby prolonging its elimination half-life. A possible explanation for our finding of lower plasma diazepam concentrations in the elderly could be that these patients have a reduced ability to absorb the drug, however there is little evidence from other studies to suggest that drug absorption is altered with age (Crooks, O'Malley & Stevenson, 1976). The influence of other compounds on the metabolism of diazepam must also be considered, especially since a number of the patients in this study were prescribed drugs which are known to increase drug metabolism, e.g. phenytoin, phenobarbitone and chlorpromazine. The mean $\pm$ s.d. age of the eighteen patients receiving multiple drug therapy was 55.1 ± 12.3 compared to 44.9 ± 13.9 years for those prescribed diazepam only and this difference was significant ($t=2.34$, $P=0.025$). Thus it is possible that the lower plasma concentrations were a result of the interaction between diazepam and other prescribed drugs. No evidence was found in this study to support the suggestion by Kanto *et al.* (1974) that diazepam induces its own metabolism.

The association between the plasma concentrations of desmethyldiazepam and diazepam was remarkably close and in nearly all the samples analysed the metabolic concentration was greater than that of the parent compound. In only one patient sample (no. 16) was the concentration of desmethyldiazepam less than that of diazepam and, in this case, it is most likely that the patient had not been taking her medication regularly. Since the relationship between these two compounds was unaffected by the duration of therapy, comparison of the plasma concentrations of both desmethyldiazepam and diazepam can be very useful when assessing patient compliance.

We thank B. Soobrayen for help with the analyses and the consultant, nursing and pharmacy staff at Knowle Hospital, Fareham, for their co-operation in obtaining plasma samples from the in-patient group.

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(Received July 18, 1977)