Br. J. clin. Pharmac. (1986), **22**, 421–428

Pharmacokinetics of the new benzodiazepine antagonist Ro 15-1788 in man following intravenous and oral administration

G. RONCARI[1], W. H. ZIEGLER[2] & T. W. GUENTERT[1]
[1]Division of Clinical Pharmacology, F. Hoffmann-La Roche & Co. Ltd, CH-4002 Basel and [2]Division of Clinical Pharmacology, Department of Medicine, University Hospital, CH-8091 Zurich, Switzerland

1 During clinical pharmacology studies with the benzodiazepine antagonist Ro 15-1788 the pharmacokinetic characteristics of high intravenous doses (20 and 40 mg) and of an oral dose (200 mg) were examined in six healthy male volunteers.
2 Ro 15-1788 was rapidly and extensively distributed in the body with an apparent volume of distribution V_{ss} of 1.06 l kg^{-1}. Elimination occurred rapidly by hepatic metabolism and the high plasma clearance of 1.14 l min^{-1} resulted in a short elimination half-life of less than 1 h.
3 No difference in the disposition parameters calculated from the data after the 20 and 40 mg doses was observed reflecting a dose-proportionality in the areas under plasma concentration-time curves and unchanged distribution characteristics.
4 Because the blood/plasma distribution coefficient is close to unity the disposition parameters obtained from plasma concentrations are similar to the corresponding parameters with reference to blood.
5 Following oral administration of 200 mg the drug is rapidly absorbed. Peak levels were reached after 20–90 min and were close to or even higher than the values after the 40 mg intravenous dose at the same time point. Due to the high hepatic extraction ratio the fraction reaching the systemic circulation unchanged was reduced to approximately 16% during the absorption step.

Keywords benzodiazepine antagonist Ro 15-1788 pharmacokinetics absorption disposition

Introduction

Ro 15-1788, an imidazobenzodiazepine derivative (Figure 1), is a new benzodiazepine antagonist (Hunkeler *et al.*, 1981; Möhler & Richards, 1981). Structurally it is related to the 1,2-annelated benzodiazepines midazolam and triazolam, both short acting hypnotics. In particular it shares the imidazo residue with midazolam. However, physico-chemical properties of the two molecules are different. Not only is the antagonist (pK_a 1.7) much less ionized at physiologic pH than midazolam (pK_a 6.1), but it is also more polar as indicated by the much lower partition coefficient in octanol-buffer pH 7.5 (14 *vs* 475). Ro 15-1788 lacks the pharmacological effects of classical benzodiazepines but it binds specifically to benzodiazepine receptors in the brain blocking in this way the behavioural, neurological and electrophysiological effects of several benzodiazepines (Darragh *et al.*, 1981, 1982; Gaillard & Blois, 1983; Klotz *et al.*, 1984a).

Correspondence: Dr G. Roncari, c/o F. Hoffmann-La Roche & Co. Ltd, Grenzacherstrasse, CH-4002 Basel, Switzerland

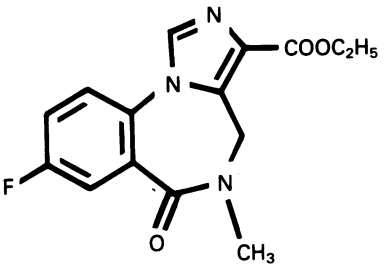

Figure 1 Molecular structure of the benzodiazepine antagonist Ro 15-1788.

The compound does not inhibit at all the effects of barbiturates, meprobamate, ethanol, GABA mimetics and opioids.

Single oral doses up to 600 mg and intravenous doses up to 100 mg were very well tolerated by healthy volunteers (Darragh *et al.*, 1983a, b). First results in benzodiazepine over-dosage showed that this drug terminated benzodiazepine-induced coma within 1–2 min following its intravenous injection. The main clinical indications will therefore be the treatment of benzodiazepine over-dosage and the reversions of benzodiazepine effects in anaesthesia.

The present study was undertaken to evaluate possible pharmacological effects of the antagonist at high doses (20 and 40 mg i.v.; 200 mg p.o.) and to determine at the same time its pharmacokinetics at these doses. The present report deals only with the aspects on absorption and disposition characteristics of the drug.

Methods

Subjects

Six healthy male subjects (for anthropometric data see Table 1) entered the study after giving their written informed consent to the programme which had been revised and approved by the Institutional Ethics Committee. Prior to entry, the physiological status of the subjects was determined by medical history, physical examination and laboratory tests of haematopoietic, hepatic and renal function. Two weeks prior to the start of the study and during the study the subjects received no other medication.

Table 1 Subject characteristics

Subject	*Age (years)*	*Weight (kg)*	*Height (cm)*
C.C.	26	74.0	182
T.H.	27	65.2	175
T.M.	27	79.3	187
B.M.	27	72.8	179
V.K.	24	79.0	186
U.W.	27	75.0	186
Mean	26.3	74.2	182
Range	24–27	65–79	175–187

Dosing

Each subject received at weekly intervals the following four treatment combinations:

A. 20 mg Ro 15-1788 i.v. together with two coated placebo tablets orally.
B. two coated 100-mg Ro 15-1788 tablets orally together with 4 ml i.v. placebo solution.
C. 4 ml i.v. placebo solution together with two placebo tablets orally.
D. 40 mg Ro 15-1788 i.v.

Effect measurements required inclusion of the placebo treatments. Parts A–C of the study were conducted in a double-blind, randomized cross-over fashion. For safety reasons the 40 mg intravenous dose was not included in the double blind procedure.

Before a drug administration the subjects fasted overnight (no food and alcoholic beverages from 22.00 h on) and drank only a glass of water before medication. For intravenous administration 4 ml (8 ml) of a water/organic solvents mixture containing 20 mg (40 mg) of Ro 15–1788 or the same volume of a placebo solution was injected into an arm vein within 1 min for the smaller and within 2 min for the larger volume. For oral administration two coated tablets containing together 200 mg Ro 15-1788 or placebo were swallowed with 100 ml of tap water.

Two hours after administration the subjects took a light breakfast with bread, marmalade and tea. During the first 6 h after the administration the subjects drank small portions of water at 30 min intervals to assure a sufficient urine flow. No other food was taken until after the 6 h blood collection.

Sample collection

Following every drug administration blood samples of 8 ml each were collected via a venous catheter with obturator into Vacutainer® tubes using K-NH_4-oxalate as anticoagulants at the following times: before administration, 5, 10, 20, 30, 45 min and 1, 1.5, 2, 2.5, 3, 4, and 6 h after administration. Within 60 min after sampling blood was centrifuged at 1000 *g*, the plasma separated and stored at −20° C until analyzed.

Total urine was collected before administration, and up to 24 h post administration. The samples were stored at −20° C until analyzed.

Drug analysis

The concentrations of drug in plasma and urine were determined by an h.p.l.c. method described earlier (Timm & Zell, 1983).

Duplicate determinations were performed with each plasma and urine sample. Precision and accuracy of the analytical method used were assessed by analyzing together with the samples from the study quality control samples spiked with the antagonist (40 ng ml^{-1} for plasma and 50 ng ml^{-1} for urine). The quality control samples were prepared by a different person than the one performing the analysis.

Blood/plasma distribution

The distribution of drug between erythrocytes and plasma was determined at spiked blood concentrations of 20 ng and 200 ng [^{14}C]-labelled Ro 15-1788 ml^{-1} after an equilibration time of 5, 15 and 30 min. Fresh pooled blood of three healthy male donors with K-NH_4-oxalate as anticoagulants was used for this experiment. Blood and plasma samples (200 µl) from the same time point were combusted in a LS Sample Oxidizer (Intertechnique, Paris; model IN 4101) and radioactivity measured in a liquid scintillation counter with external standard (Isocap 300, Nuclear Chicago). The distribution coefficient λ was calculated according to:

$$\lambda = \frac{\text{drug concentration in blood}}{\text{drug concentration in plasma}}$$

Data analysis

The evaluation of pharmacokinetic parameters was performed in two ways.

(1) By model-independent analysis.

Elimination rate constants (λ_z) were estimated by linear regression analysis of the terminal parts of the log-linear plasma concentration-time profiles. The elimination half-lives ($t_{½,z}$) were then obtained by dividing ln2 by λ_z. The areas under the plasma concentration-time curves ($AUC_{0-\infty}$) were calculated using the linear trapezoidal rule from time zero to the last measured concentration point and adding the residual area obtained by multiplying the last concentration value with $1/\lambda_z$. Total plasma drug clearance (CL_p) was estimated using the expression Dose/$AUC_{0-\infty}$. From this value the corresponding blood clearance (CL_b) was derived using the determined blood/plasma distribution coefficient ($CL_b = CL_p/\lambda$). An estimation of the distribution volume was obtained by calculating V_{ss} by the method proposed by Benet & Galeazzi (1979).

(2) By model-dependent analysis of the concentration data.

Measured intravenous and oral plasma concentration-time data were fitted to one or two-compartment models (for oral data with and without lag time) using nonlinear regression analysis. With the aid of the coefficients and exponents of the fitted functions the following pharmacokinetic parameters were calculated according to standard procedures (Gibaldi & Perrier, 1982): half-life of elimination ($t_{½,z}$), area under the curve ($AUC_{0-\infty}$), total plasma drug clearance (CL_p), volume of distribution of the central compartment (V_c), volume of distribution during the post-distributive phase (V).

Results

Analytical

From the results of the quality control samples ($n = 72$) determined together with the samples from the study the accuracy of the h.p.l.c. method used was found to be 0.8–2% and the precision 3.5–4%.

Pharmacokinetics

Representative examples of the plasma concentration-time curves following intravenous and oral administration of the benzodiazepine antagonist, Ro 15–1788, are given in Figure 2 (subjects C.C. and V.K.). No drug was detectable in the plasma samples collected during the placebo experiment. Following *intravenous* administration the drug distributed extensively outside the vascular space as was indicated by average volumes of distribution (V_{ss}) of over 1 l kg^{-1} (1.11 l kg^{-1} after 20 mg; 1.01 l kg^{-1} after 40 mg dose; see Table 2). The distribution process was rapidly concluded and only in four out of six subjects a distribution phase could be discerned in the plasma concentration-time profiles, although the first blood sample was collected in all cases as early as 5 min after dosage.

No preferential distribution of drug into red blood cells occurred. The blood/plasma partition coefficient was on average 0.88. The values were identical at all three equilibration times chosen indicating that the equilibrium was reached already before the first observation time at 5 min. No difference between the values

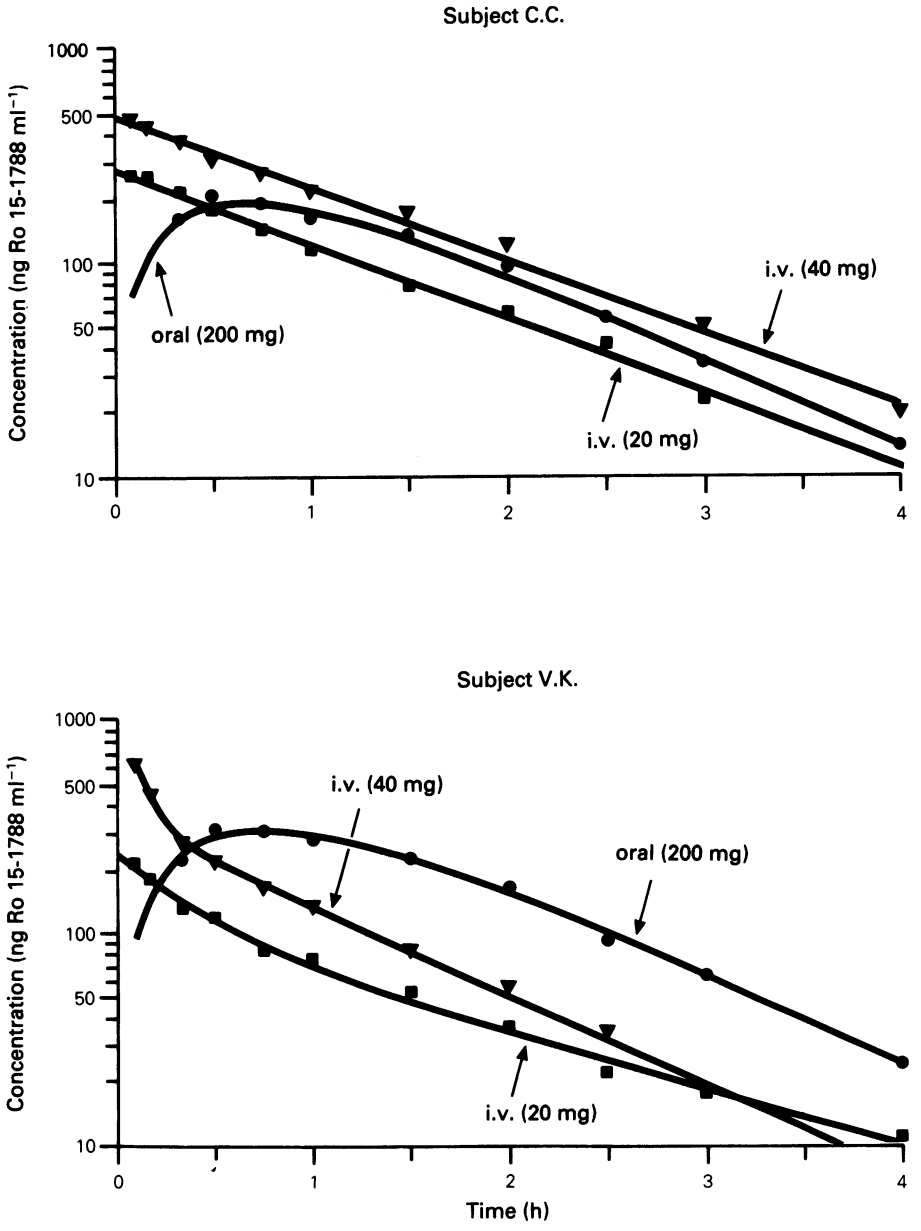

Figure 2 Plasma concentration-time curves of Ro 15-1788 following three different Ro 15-1788 doses to two healthy male volunteers.

obtained at 20 ng ml^{-1} (mean ± s.d.: 0.87 ± 0.01) and at 200 ng ml^{-1} (0.89 ± 0.01) was found.

Shortly after the intravenous administration plasma concentrations declined log-linearly suggesting that the elimination of the benzodiazepine antagonist proceeds by a first-order process. Elimination half-lives around 50–60 min were estimated by log-linear regression analysis (Table 2). After both doses given the drug was rapidly cleared from plasma. Clearance values averaged at 14.8 ml min^{-1} kg^{-1} for the 20 mg dose and at 16.2 ml min^{-1} kg^{-1} for the 40 mg dose (Table 2). Since the blood/plasma distribution ratio was

Table 2 Disposition parameters of Ro 15-1788 obtained in a model-independent way

	20 mg dose				*40 mg dose*			
Subject	$AUC_{o-\infty}$ (ng ml^{-1} h)	CL_p (ml min^{-1} kg^{-1})	V_{ss} (l kg^{-1})	$t_{½,z}$ (min)	$AUC_{o-\infty}$ (ng ml^{-1} h)	CL_p (ml min^{-1} kg^{-1})	V_{ss} (l kg^{-1})	$t_{½,z}$ (min)
C.C.	340	13.2	0.97	52	646	13.9	1.08	54
T.H.	347	14.7	1.30	69	551	18.6	1.16	52
T.M.	260	16.1	1.09	55	500	16.8	0.97	47
B.M.	373	12.2	0.94	57	584	15.7	1.13	57
V.K.	236	17.8	1.47	63	428	19.7	0.99	43
U.W.	326	13.6	0.88	45	724	12.3	0.70	39
Mean	314	14.8	1.11	57	572	16.2	1.01	49
s.d.	54	2.30	0.23	8	105	2.80	0.17	7

found to be close to unity these values reflect closely the respective blood clearances.

A paired *t*-test revealed no difference (α = 0.05) in the disposition parameters obtained at the two dosages given to the same volunteers. Pooled parameters (mean ± s.d.), therefore, may be used to describe the disposition of the drug (CL_p: 15.5 ± 2.5 ml min^{-1} kg^{-1}; V_{ss}: 1.06 ± 0.20 l kg^{-1}; $t_{½,z}$: 53 ± 8.5 min).

Disposition parameters obtained from compartmental analysis (Table 3) were practically identical with those calculated in a more model-independent way. The good agreement confirms the adequacy of the chosen models.

Following *oral* administration of 200 mg Ro 15-1788 peak plasma concentrations of 143 to 439 ng ml^{-1} were reached within 20 to 45 min with one exception (subject T.H. = 90 min; Table 4). After the peak levels were reached, the plasma concentrations declined rapidly and were below the detection limit of the h.p.l.c. method used (10 ng ml^{-1}) 4–6 h after administration. In five of the six subjects studied enough data points were available to define the slope of the log-linear terminal phase of the concentration curves. Half-life values in the range of 42–71 min (mean: 49 min) were observed (Table 4). These values were in good agreement with the corresponding parameters obtained by compartmental modeling of the oral concentration-time curves. However, due to the limited number of blood samples available after oral dosing the data of only four subjects did lend themselves for a model-dependent analysis.

Based on area comparisons the bioavailability of Ro 15-1788 from the oral dosage form employed could be calculated. Little difference was seen in this estimate when the data from the 20-mg intravenous dose (F = 15 ± 7%) or when those of the 40 mg dose (F = 17 ± 8%) were taken as standard of reference (Table 4).

Discussion

In spite of its low lipophilicity Ro 15-1788 has a large apparent volume of distribution comparable to that of other, more lipophilic benzodiazepines (for review see Guentert, 1984). The steady-state volume of distribution is with 1.06 l kg^{-1} similar to the values for midazolam (0.7 l kg^{-1}; Allonen *et al.*, 1981; Amrein *et al.*, 1981) and triazolam (1.1 l kg^{-1}; Eberts *et al.*, 1981) indicating that the antagonist is distributed outside the plasma and bound to tissue components. This unexpected finding of a high volume of distribution may be explained, at least in part, by the relatively low binding of Ro 15-1788 to plasma constituents. While most therapeutically used benzodiazepines are over 90% bound to albumin (Guentert, 1984), the bound fraction of the antagonist amounts only to 40–50% (Klotz *et al.*, 1984b). No preferential distribution of drug into red blood cells occurs. The blood/plasma partition coefficient determined using pooled blood from three healthy volunteers was 0.88 which was in close agreement with the mean value of 0.99 reported by Klotz *et al.* (1984b).

Distribution of the antagonist in the body is rapid as indicated by only short distribution phases in plasma concentration-time profiles. The contribution of the distribution phase (λ_1-phase) to the overall area under the plasma concentration-time curve was in all four cases small (< 20%). This rapid distribution explains the quick onset of the antagonist effect reversing benzodiazepine-induced sedation within 1–2 min following injection (Geller *et al.*, 1985).

Ro 15-1788 is rapidly cleared from the body. Less than 0.1% of a dose is excreted unchanged in urine. Based on filtration alone, a renal clearance of 60–70 ml min^{-1} (product of free fraction and glomerular filtration rate; Rowland & Tozer, 1980) would be expected, a value much higher

Table 3 Disposition parameters of Ro 15-1788 obtained by compartmental analysis

Dose	20 mg							40 mg						
Subject	C.C.	T.H.	T.M.	B.M.	V.K.	U.W.	Mean ± s.d.	C.C.	T.H.	T.M.	B.M.	V.K.	U.W.	Mean ± s.d.
Model (number of compartments)	1	2	2	2	2	1		1	2	2	2	2	1	
A (ng ml^{-1})	–	269	250	181	118	–	205 ± 69.0	–	918	713	639	735	–	751 ± 119
B (ng ml^{-1})	–	199	170	254	112	–	184 ± 59.2	–	366	385	393	342	–	372 ± 22.7
λ_1 (h^{-1})	–	9.86	6.61	8.33	2.69	–	6.87 ± 3.09	–	10.5	12.0	12.3	10.3	–	11.3 ± 1.02
λ_z (h^{-1})	0.801	0.625	0.774	0.732	0.601	0.916	0.742 ± 0.117	0.773	0.799	0.871	0.743	0.951	1.05	0.865 ± 0.118
$AUC_{0-\infty}$ (ng ml^{-1} h)	340	346	258	369	231	322	311 ± 54	627	546	502	581	430	710	566 ± 98
Cl_p (ml min^{-1} kg^{-1})	13.2	14.7	16.3	12.4	18.2	13.9	14.8 ± 2.14	14.3	18.7	16.8	15.8	19.6	12.5	16.3 ± 2.66
V_c (l kg^{-1})	0.99	0.65	0.60	0.63	1.10	0.90	0.81 ± 0.21	1.11	0.48	0.46	0.53	0.47	0.71	0.63 ± 0.25
V (l kg^{-1})	–	1.42	1.26	1.02	1.82	–	1.38 ± 0.34	–	1.41	1.15	1.27	1.24	–	1.27 ± 0.11

than the approximately 1 ml min^{-1} observed. This low value indicates that Ro 15-1788 is extensively reabsorbed in the kidney tubule. The entire plasma clearance can therefore be attributed to metabolic degradation. The high clearance results in a rapid decline of drug in the body. Elimination half-life values of less than 1 h were observed. The rapid disappearance of the antagonist from the body after a single dose may lead to recurrence of drowsiness when reversal of sedation after a long-acting benzodiazepine is attempted. This situation may therefore require administration of a prolonged infusion or multiple intravenous doses of the antagonist.

In the present investigation every subject received two different intravenous drug doses allowing to test for possible changes in the disposition parameters with dose. There was good intraindividual agreement in the model used to describe the disposition as well as in the clearance, volume of distribution and elimination half-life values obtained after the two doses administered. It can therefore be concluded that the disposition of Ro 15-1788 is independent of the concentration at least within the limited dose range examined. The observation further points to a small intraindividual variability in the disposition of the drug. This variability in the parameters expressed by the average of all individuals' coefficient of variation (CV) (CL_p: 9.5%; V_{ss}: 13.4%; $t_{½,z}$: 11.7%) was similar to the interindividual variability (CL_p 15%; V_{ss} 15%; $t_{½,z}$ 12%). The small variability in the parameter values between subjects may at least in part be a consequence of the homogenous group of subjects selected for the study.

The results from the present study agree well with the data reported by Klotz *et al.* (1985) studying the kinetics of the antagonist at a dose of 0.1 mg kg^{-1} in presence or absence of flunitrazepam and lormetazepam (interaction study). In a previous communication (Klotz *et al.*, 1984b), however, the same group of investigators had reported substantially lower mean values for clearance (691 ml min^{-1}) and volume of distribution V (0.82 l kg^{-1}) than found in their later interaction study (1114 ml min^{-1}; 1.53 l kg^{-1}). The good agreement in the values from our present study obtained after 20 and 40 mg doses and the interaction study by Klotz *et al.* (1985) after doses around 7 mg suggests that not differences in dosage but rather in sampled populations may explain the discrepancy in results.

Although Ro 15-1788 was administered orally in several efficacy studies (Darragh *et al.*, 1982; Lupulover *et al.*, 1984) no data is given in the literature about the absorption characteristics of the drug following oral dosage. The present

Table 4 Pharmacokinetic parameters of Ro 15-1788 following oral administration

	Subject						
	C.C.	*T.H.*	*T.M.*	*B.M.*	*V.K.*	*U.W.*	*Mean ± s.d.*
C_{max} (ng ml^{-1})	210	143	439	282	308	147	255 ± 113
t_{max} (min)	30	90	20	30	30	45	41 ± 25
$AUC_{0-\infty}$ (ng ml^{-1} h)	382	308	429	656	632	243	442 ± 169
$t_{½,z}$ (min)	43	—*	46	71	45	42	49 ± 12
Bioavailability i.v. reference							
20 mg	11%	9%	17%	18%	27%	7%	15 ± 7%
40 mg	12%	11%	17%	22%	30%	7%	17 ± 8%

*Not enough data points available to characterize the elimination phase. For extrapolation of the area the slope defined by the last three data points was employed.

study included therefore an oral administration. Following administration of two 100 mg tablets the absorption of the drug was rapid. Average (± s.d.) peak plasma concentrations of 255 (± 113) ng ml^{-1} were reached in all but one case in less than 1 h (range: 20–90 min). The half-life of the absorption phase averaged at 0.27 h again indicating that the absorption process was completed 1 h post administration. Around 1 h after oral dosage plasma levels were close to or even exceeded those seen after the 40 mg intravenous dose. Although previous mass balance studies had indicated that all of orally administered drug was taken up into the portal blood (Wendt *et al.*, personal communication), only around 16% of an oral dose reached the systemic circulation unchanged regardless of which one of the two intravenous doses was taken as standard of comparison. Considering the high hepatic clearance this reduction in the bioavailability is not unexpected. The close correspondence of total blood clearance (approximately 1.3 l min^{-1}) and hepatic blood flow (around 1.5 l min^{-1}) is suggestive of a large extraction ratio of the drug in the liver. Applying flow model considerations the bioavailability can be predicted based on the drug's disposition characteristics (Rowland, 1972). Using these principles the average predicted bioavailability of 19% corresponds well with the experimentally obtained average value. This would indicate that the low availability is only due to a first-pass effect.

Conclusions

From our studies it can be concluded that Ro 15-1788 like other 1,2-annelated benzodiazepines is rapidly and extensively distributed in the body assuring rapid onset of effects. Elimination occurs rapidly by hepatic metabolism and the high plasma clearance of 1.14 l min^{-1} results in a short elimination half-life of less than 1 h and hence in a short duration of reversal of sedative benzodiazepine effects. No difference in the disposition parameters calculated from the data after the 20 and 40 mg doses was observed reflecting dose-proportionality in the areas under the plasma concentration-time curves and unchanged distribution characteristics after the two different doses. Following oral administration of 200 mg the drug is rapidly absorbed with peak levels reached after 20–90 min close to or even higher than the values after the 40 mg intravenous dose at the same time point. Due to the high hepatic extraction ratio orally administered drug undergoes pronounced first-pass metabolism reducing the dose fraction reaching the systemic circulation unchanged to approximately 16%.

References

Allonen, H., Ziegler, G. & Klotz, U. (1981). Midazolam kinetics. *Clin. Pharmac. Ther.*, **30**, 653–661.

Amrein, R., Cano, J. P., Eckert, M. & Coassolo, P. (1981). Pharmakokinetik von Midazolam nach intravenöser Verabreichung. *Arzneim.-Forsch. (Drug Res.)*, **31**, 2202–2205.

Benet, L. Z. & Galeazzi, R. L. (1979). Noncompartmental determination of the steady-state volume of distribution. *J. pharm. Sci.*, **68**, 1071–1074.

Darragh, A., Lambe, R., Kenny, M. & Brick, I. (1983b). Tolerance of healthy volunteers to intravenous administration of the benzodiazepine antagonist Ro 15-1788. *Eur. J. clin. Pharmac.*, **24**, 569–570.

Darragh, A., Lambe, R., Kenny, M., Brick, I., Taaffe, W. & O'Boyle, C. (1982). Ro 15-1788 antagonises

the central effects of diazepam in man without altering diazepam bioavailability. *Br. J. clin. Pharmac.*, **15**, 529–536.

Darragh, A., Lambe, R., O'Boyle, C., Kenny, M. & Brick, I. (1983a). Absence of central effects in man of the benzodiazepine antagonist Ro 15-1788. *Psychopharmacology*, **80**, 192–195.

Darragh, A., Scully, M., Lambe, R., Brick, I., O'Boyle, C. & Downie, W. W. (1981). Investigation in man of the efficacy of a benzodiazepine antagonist, Ro 15-1788. *Lancet*, **ii**, 8–10.

Eberts, F. S., Philopoulos, Y., Reineke, L. M. & Vliek, R. W. (1981). Triazolam disposition. *Clin. Pharmac. Ther.*, **29**, 81–93.

Gaillard, J.-M. & Blois, R. (1983). Effect of the benzodiazepine antagonist Ro 15-1788 on flunitrazepam induced sleep changes. *Br. J. clin. Pharmac.*, **15**, 529–536.

Geller, E., Niv, D., Silbiger, A., Halpern, P., Leykin, Y., Rudick, V. & Sorkin, P. (1985). Ro 15-1788. A benzodiazepine antagonist in the treatment of 34 intoxicated patients. *Anesthesiol.*, **63**, A157.

Gibaldi, M. & Perrier, D. (1982). *Pharmacokinetics*. New York: Marcel Dekker Inc.

Guentert, T. W. (1984). Pharmacokinetics of benzodiazepines and of their metabolites. In *Progress in Drug Metabolism*. Vol. 8, eds Bridges, J. W. & Chasseaud, L. F. pp. 241–386. London and Philadelphia: Taylor and Francis.

Hunkeler, W., Möhler, H., Pieri, L., Polc, P., Bonetti, E. P., Cumin, R., Schaffner, R. & Haefely, W. (1981). Selective antagonists of benzodiazepines. *Nature*, **290**, 514–516.

Klotz, U., Duka, Th., Dorow, R. & Doenicke, A. (1985). Flunitrazepam and lormetazepam do not affect the pharmacokinetics of the benzodiazepine antagonist Ro 15-1788. *Br. J. clin. Pharmac.*, **19**, 95–98.

Klotz, U., Ludwig, L. & Ziegler, G. (1984a). Midazolam and benzodiazepine-antagonists: Pharmacokinetic and pharmacodynamic interactions in man. *Naunyn-Schmiedeberg's Arch. Pharmac.*, **325**, R 90.

Klotz, U., Ziegler, G. & Reimann, I. W. (1984b). Pharmacokinetics of the selective benzodiazepine antagonist Ro 15-1788 in man. *Eur. J. clin. Pharmac.*, **27**, 115–117.

Lupulover, Y., Safran, A. B., Desangles, D., De Weisse, C., Meyer, J. J., Bousquet, A. & Assimacopoulos, A. A. (1984). Evaluation of visual function in healthy subjects after administration of Ro 15-1788. *Eur. J. clin. Pharmac.*, **27**, 505–507.

Möhler, H. & Richards, J. G. (1981). Agonist and antagonist benzodiazepine receptor interaction *in vitro*. *Nature*, **294**, 763–765.

Rowland, M. (1972). Influence of route of administration on drug availability. *J. pharm. Sci.*, **67**, 70–74.

Rowland, M. & Tozer, T. N. (1980). *Clinical pharmacokinetics: Concepts and applications*. Philadelphia: Lea & Febiger.

Timm, U. & Zell, M. (1983). Determination of the benzodiazepine antagonist Ro 15-1788 in plasma by high-performance liquid chromatography with UV detection. *Arzneim.-Forsch. (Drug Res.)*, **33**, 358–362.

(Received 4 November 1985, accepted 11 June 1986)