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Flunitrazepam and lormetazepam do not affect the pharmacokinetics of the benzodiazepine antagonist Ro 15–1788

U. KLOTZ[1], Th. DUKA[2], R. DOROW[3] & A. DOENICKE[2]
[1]Dr Margarete Fischer-Bosch-Institut für Klinische Pharmakologie Stuttgart, [2]Abteilung für experimentelle Anaesthesie der Chirurgischen Klinik und Poliklinik Innenstadt der Universität München, and [3]Forschungslaboratorien der Schering AG Berlin (West)/Bergkamen, FRG

Following a single intravenous dose of 0.1 mg/kg, the pharmacokinetics of Ro 15-1788, a specific benzodiazepine antagonist, have been investigated in 20 healthy male volunteers. In random order injection of Ro 15-1788 has been preceded (5 min) by intravenous dosing with 0.06 mg/kg of lormetazepam (n=6), 0.03 mg/kg of flunitrazepam (n=8) or placebo (n=6). The rapid elimination of the antagonist could be characterized by an elimination half-life between 0.9 to 1.4 h and a total plasma clearance of 727 to 1440 ml/min. These single dose studies indicate that the disposition of Ro 15-1788 was not affected by the acute coadministration of both benzodiazepines.

Keywords pharmacokinetics benzodiazepines Ro 15-1788

Introduction

Benzodiazepine derivates suitable for induction of general anaesthesia must possess a short onset of action (induction times not longer than 1–2 min; for review see Kanto & Klotz, 1982). Following i.v. administration of diazepam or flunitrazepam sleep is induced in such an acceptable period of time, with flunitrazepam acting slightly faster than diazepam (Stovner *et al.*, 1973). Apparently the new hypnotic midazolam acts even more rapidly (Allonen *et al.*, 1981) and intravenous lormetazepam might be also a candidate for induction of anaesthesia (Doenicke *et al.*, 1979).

Recently different compounds have been tried to antagonize the sedative hypnotic effects of benzodiazepines. These attempts might be clinically helpful, if recovery time or residual effects have to be minimized. Conflicting data have been reported for physostigmine (Avant *et al.*, 1979; Grote *et al.*, 1981; Pandit *et al.*, 1983) and the observed effects of theophylline (Arvidsson *et al.*, 1982; Henauer *et al.*, 1983) might be due to ethylenediamine which is part of the aminophylline preparation used and which is known to interact with the binding of [^{3}H]-diazepam in cerebral cortical synaptosomal membranes (Morgan & Stone, 1983). The new imidazodiazepine Ro 15-1788 can reverse the sedative effects of 3-methylclonazepam (Darragh *et al.*, 1981), diazepam (Darragh *et al.*, 1982), flunitrazepam (Gaillard & Blois, 1983) and midazolam (Klotz *et al.*, 1984a).

Since the antagonistic effects are short-lasting, a rapid elimination of Ro 15-1788 has to be assumed, which was substantiated very recently (Klotz *et al.*, 1984b). Otherwise, so far limited data are available for the pharmacokinetics of Ro 15–1788, especially whether the coadministered benzodiazepines can influence the disposition of the selective benzodiazepine antagonist. Therefore we evaluated in a placebo-controlled trial, whether flunitrazepam or lormetazepam affected the pharmacokinetics of Ro 15–1788.

Correspondence: Prof. Dr U. Klotz, Dr Margarete Fischer-Bosch, Institut für Klinische Pharmakologie, D-7000 Stuttgart-50, Auerbachstrasse 112, FRG.

Methods

Clinical protocol:

After written informed consent was obtained 20 healthy drug-free male volunteers (age range: 18–36 years, weight: 59–87 kg) were randomly assigned to placebo- (n = 6), lormetazepam- (single i.v. dose of 0.06 mg/kg; n = 6) or flunitrazepam- (single i.v. dose of 0.03 mg/kg; n = 8) treatment. These doses have been proven as useful in our clinic for premedication and/or inducing anaesthesia. Five minutes after the injection (1 min) of the benzodiazepine or placebo Ro 15–1788 was administered intravenously (0.1 mg/kg) over 1 min. Blood samples were collected prior to and 1, 5, 15, 30, 45 min, 1, 1.25, 1.75, 2.25, 2.75, 3.75 and 5.75 h following the injection of Ro 15–1788. The centrifuged serum samples were stored frozen (−25°C) until analysis.

Analytical and pharmacokinetic measurements

Serum levels of Ro 15-1788 were monitored by a specific and sensitive h.p.l.c.-assay (Timm & Zell, 1983) with some slight modifications (Klotz *et al.*, 1984b). Ether extracts of plasma (+ internal standard Ro 15-6166) were separated on a reversed phase column with the mobile phase 60% 0.05 M phosphate buffer pH2.5, 35% methanol and 5% acetonitrile. The coefficients of variation of the assay were 4% (intra-assay precision) and 5.3% (between run precision), respectively. The lower limit of sensitivity was 2 ng/ml. Elimination half-life ($t_{1/2}$) was calculated from the terminal slope (0.25 to 5.75 h postdosing) by linear regression analysis, total body clearance (CL) from the ratio dose/area under the curve (AUC) and the apparent volume of distribution (V) by the area method. Statistical comparison was performed by ANOVA; all values given represent mean ± s.d.

Results

Following the bolus injection of Ro 15–1788 (0.1 mg/kg) serum concentrations declined mono-exponentially after 15 min and the time profile was not altered by the pre-injection of flunitrazepam or lormetazepam (Figure 1). The individually derived pharmacokinetic parameters for the antagonist's elimination are summarized in Table 1; again no differences were observed between the three groups studied. Values for $t_{1/2}$ and CL ranged between 0.9 to 1.4 h and 727 to 1440 ml/min, respectively.

Discussion

Single oral doses of Ro 15–1788 (200 mg) effectively antagonize the sedative-hypnotic effects of a single oral dose of 8 mg 3-methylclonazepam and of 40 mg diazepam, respectively (Darragh *et al.*, 1981, 1982). Similarly, 100 mg of the antagonist (p.o.) reversed the hypnogenic effect of 2 mg of oral flunitrazepam (Gaillard & Blois, 1983). The hypnotic action of midazolam steady state infusions

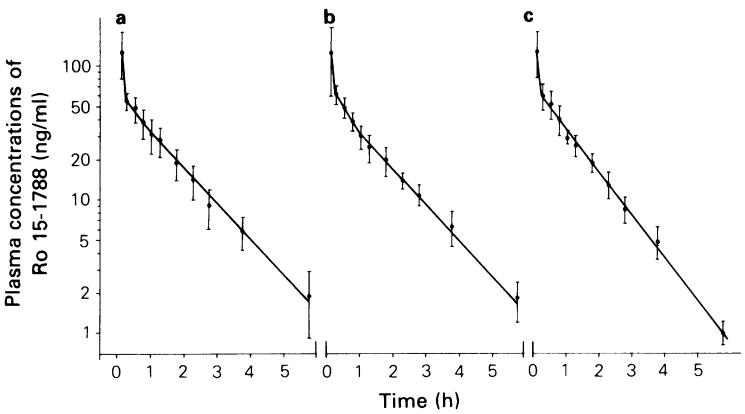

Figure 1 Plasma concentration time profiles (mean ± s.d.) of Ro 15-1788 following a single intravenous dose of 0.1 mg/kg.
(a) an i.v. bolus of 0.03 mg/kg flunitrazepam was injected 5 min prior to the benzodiazepine antagonist, (b) saline injection preceded the bolus Ro 15-1788 and (c) an i.v. bolus of 0.06 mg/kg lormetazepam was given 5 min prior to the benzodiazepine antagonist.

Table 1 Elimination of Ro 15–1788 following a single i.v. dose of 0.1 mg/kg without and with pre-injection of flunitrazepam or lormetazepam (mean ± s.d.)

	+ flunitrazepam (n = 8)	*+ placebo* (n = 6)	*+ lormetazepam* (n = 6)
$t_{1/2}$ (h)	1.13 ± 0.12	1.15 ± 0.14	0.97 ± 0.13
CL (ml/min)	1211 ± 303	1114 ± 207	1038 ± 241
V (l/kg)	1.55 ± 0.30	1.53 ± 0.43	1.23 ± 0.18

(0.025–0.04 mg kg^{-1} h^{-1}) can be antagonized by an intravenous bolus of 2.5 mg Ro 15-1788 (Klotz *et al.*, 1984a). With the exception of the last study in these clinical trials plasma levels of Ro 15–1788 have not been monitored and a relatively fast elimination has been assumed from its shortlasting action. Our results indicate, that independent of the acute coadministration of benzodiazepines, a single dose of Ro 15–1788 is rapidly eliminated with a $t_{1/2}$ of about 1 h. The high CL-value of approximately 1200 ml/min suggests a blood flow dependent type of hepatic elimination. In a recent case report diazepam-induced prolonged coma could be reversed for about 2 h following multiple dosing with Ro 15–1788 of 2.5 or 5 mg (Rapold *et al.*, 1984). This would approximate an infusion rate (R_o) of 1.5 mg/h for an effective antagonistic effect and at steady state R_o should equal the CL-rate of Ro 15–1788. Thus according to the equation

$$R_o = C_{ss} \cdot CL \text{ and } C_{ss} = \frac{R_o}{CL} = \frac{25 \text{ ug/min}}{1200 \text{ ml/min}}$$

steady state concentrations (C_{ss}) of Ro 15–1788 in the range of 20 ng/ml would be necessary for its antagonistic potency. These indirect assumptions are in excellent agreement with our own data from a double-blind study with steady state infusion of midazolam (0.025–0.04 mg kg^{-1} h^{-1}) and a single i.v. bolus of Ro 15–1788 (2.5 mg) where plasma levels between 60 and 10 ng/ml were able to reverse midazolam-induced hypnosis which was verified by EEG-recordings and psychometric tests (Klotz *et al.*, 1984a).

Based on clinical evidence and our pharmacokinetic data derived from the applied single dose the following dosage regimen for the benzodiazepine antagonist Ro 15–1788 could be suggested: a bolus of 2.5 mg is sufficient to reverse abruptly (within 1 min) benzodiazepine-induced hypnosis; subsequently an infusion rate of 25 μg/min would maintain antagonistic plasma levels; duration of infusion depends on the rate of elimination of the benzodiazepine applied. Thus, both Ro 15–1788 and the knowledge of its pharmacokinetics are valuable aids for the therapeutic use of Ro 15–1788 in reversing benzodiazepine-induced central nervous depression. Obvious clinical applications of Ro 15–1788 will be in anaesthesiology as an antidote in case of benzodiazepine overdosage and in other clinical situations (e.g. endoscopy) to abbreviate recovery time and to prevent sedative residual effects of the commonly used benzodiazepines.

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References

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

Arvidsson, S. B., Ekström-Jodal, B., Martinell, S. A. G. & Niemand, D. (1982). Aminophylline antagonises diazepam sedation. *Lancet*, **ii**, 1467.

Avant, G. R., Speeg, Jr K. V., Freemon, F. R., Schenker, S. & Berman, M. L. (1979). Physostigmine reversal of diazepam-induced hypnosis. *Ann. Intern. Med.*, **91**, 53–55.

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

Darragh, A., Lambe, R., Kenny, M., Brick, I., Taaffe, W. & O'Boyle, C. (1982). Ro 15–1788 antagonises the effects of diazepam in man without altering diazepam bioavailability. *Br. J. clin. Pharmac.*, **14**, 677–682.

Doenicke, A., Kugler, J., Kropp, M., Laub, M. & Kalbfleisch, G. (1979). Der hypnotische Effekt des neuen Benzodiazepinderivates Lormetazepam. *Anaesthesist*, **28**, 578–583.

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.

Grote, B., Doenicke, A., Kugler, J., Laub, M., Ott, H., Fichte, K., Suttmann, J. & Zwisler, P. (1981). Die antagonistische Wirkung von Physostigmin auf die Sedierung durch Lormetazepam. *Anaesthesist,* **30**, 627–632.

Henauer, S. A., Hollister, L. E., Gillespie, H. K. & Moore, F. (1983). Theophylline antagonizes diazepam-induced psychomotor impairment. *Eur. J. clin. Pharmac.,* **25**, 743–747.

Kanto, J. & Klotz, U. (1982). Intravenous benzodiazepines as anaesthetic agents: Pharmacokinetics and clinical consequences. *Acta Anaesth. Scand.,* **26**, 554–569.

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.

Morgan, P. F. & Stone, T. W. (1983). Structure-activity studies on the potentiation of the benzodiazepine receptor binding by ethylendiamine analogues and derivatives. *Br. J. Pharmac.,* **79**, 973–978.

Pandit, U. A., Kothary, S. P., Samara, S. K., Domino, E. F. & Pandit, S. K. (1983). Physostigmine fails to reverse clinical, psychomotor, or EEG effects of lorazepam. *Anesth. Analg.,* **62**, 679–685.

Rapold, H. J. Follath, F., Scollo-Lavizzari, G, Kehl, O. & Ritz, R. (1984). Verlängertes Koma durch Sedation mit Diazepam bei beatmeten Patienten *Dtsch Med. Wschr.,* **109**, 340–344.

Stovner, J., Endresen, R. & Österud, A, (1973). Intravenous anaesthesia with a new benzodiazepine Ro 14–4200. *Acta Anaesth. Scand.* **17**, 163–168.

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.

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