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Lack of an Effect of Madopar on the Disposition of Tolcapone and its 3-*O*-Methylated Metabolite in Rats

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Abstract

The effect of Madopar (benserazide and L-dopa, 1:4) on the disposition of the new selective inhibitor of catechol-*O*-methyltransferase, tolcapone, in rats was investigated. There was no statistically significant difference in the pharmacokinetic parameters of tolcapone in the presence or absence of Madopar except for a change in the mean residence time after oral administration. Thus, we rejected the hypothesis that the consumption of *S*-adenyl-L-methionine by Madopar would change the disposition of tolcapone.

There were no statistically significant differences in the cumulative amount absorbed of drug and the absorption rate in the presence or absence of Madopar. We concluded that there was no interaction between tolcapone and Madopar.

The combination of Madopar (benserazide and L-dopa, 1:4) or Sinemet (carbidopa and L-dopa, 1:10 and 1:4) with a catechol-*O*-methyltransferase (COMT) inhibitor improves the relative bioavailability of L-dopa by reducing the conversion of L-dopa to 3-*O*-methyldopa (Kaakkola et al 1990; Zürcher et al 1990). Tolcapone (Ro 40-7592: 3,4-dihydroxy-4'-methyl-5-nitrobenzophenone, a new selective inhibitor of COMT (Zürcher et al 1990)) in combination with Madopar is presently being tested in clinical trials for Parkinson's disease. In general, when the decarboxylation route of L-dopa is blocked, L-dopa is effectively metabolized by COMT (Rose et al 1991) with the consumption of *S*-adenyl-L-methionine (SAM). Since SAM is a cofactor of COMT (Jeffery & Roth 1985), the consumption of SAM by Madopar would change the disposition of tolcapone by reducing the formation of 3-*O*-methylated tolcapone. In addition, since tolcapone is a substrate of COMT, it may be expected that co-administration of Madopar and tolcapone would affect the disposition of tolcapone.

Rabey et al (1989) reported that peak plasma levels of bromocriptine were reduced by L-dopa in Parkinson's disease and a smoothing of the plasma bromocriptine curve enables a more stable penetration of the medication into the central nervous system. On the other hand, L-dopa is absorbed by the active transport system which is normally responsible for the absorption of large, neutral amino acids (Lennernäs et al 1993). Since the cell membrane binding form of COMT is abundantly formed in the intestinal muscle layer (Nissinen et al 1988), some interaction between Madopar and tolcapone may be also anticipated during the absorption process of tolcapone. Therefore, this study aims to clarify the effect of Madopar on the disposition of tolcapone in rats.

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Materials and Methods

Materials

Tolcapone, its 3-*O*-methylated metabolite (Ro 40-7591; 3-*O*-methyl-4-hydroxy-4'-methyl-5-nitrobenzophenone), Ro 40-6031 (3,4-dihydroxy-4'-chloro-5-nitrobenzophenone; internal standard used for HPLC analysis), benserazide and L-dopa were obtained from Hoffman-La Roche (Basle, Switzerland). All other chemicals were reagent grade.

Animal experiments

Male Sprague-Dawley rats, 195–215 g (7 weeks of age), were placed in four groups of four animals each. Each group received tolcapone either 10 mg kg^{-1} intravenously or 20 mg kg^{-1} orally with or without Madopar. The rats were fasted overnight but had free access to water. Food was given 4 h after administration of the drug. For the intravenous study, Madopar (L-dopa 16 mg kg^{-1} and benserazide 4 mg kg^{-1}) was dissolved in 0·01 M HCl, pH 3·0 adjusted by 1 M HCl. The solution was administered orally at a volume of 5 mL kg^{-1}, 30 min before the administration of tolcapone to observe the intrinsic effect (i.e. systemic effect) of Madopar on the disposition of tolcapone. Tolcapone was dissolved in 60% polyethylene glycol 200 in saline (0·9% NaCl) and administered by bolus injection at a volume of 1 mL kg^{-1} into the penis vein under light ether anaesthesia.

For the oral study, a suspension of tolcapone or that of tolcapone and Madopar was made using a vehicle comprising 0·5% carmellose sodium salt, 0·4% polyoxyethylene (20) sorbitan mono-oleate and 0·5% benzyl alcohol in saline. The suspension of tolcapone or that of tolcapone and Madopar was administered via a tube into the stomach at a volume of 5 mL kg^{-1}. Co-administration of tolcapone and Madopar was performed in the oral study to study the interaction during the absorption process, since the clinical dose would be taken the same way.

Each rat was held in a separate metabolic cage after the above administration and blood samples (100 μL) were

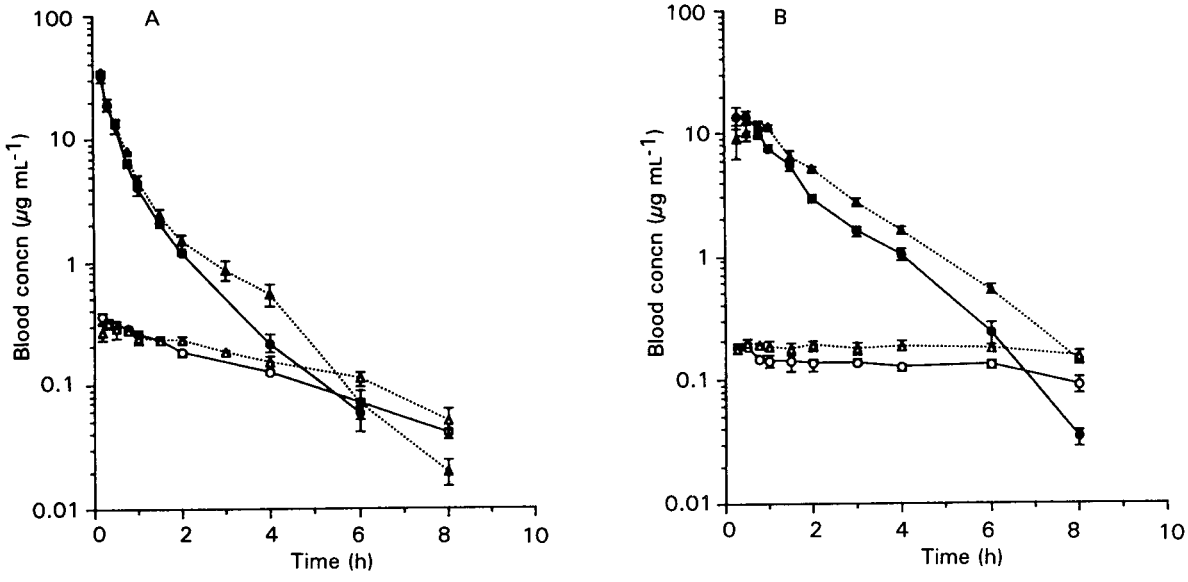

FIG. 1. The blood concentration–time course of tolcapone (●, ▲) and its 3-*O*-methylated metabolite (○, △) following intravenous administration at dose of 10 mg kg^{-1} (A) and following oral administration at a dose of 20 mg kg^{-1} (B), without Madopar (●, ○) and with Madopar (▲, △). Data show the mean ± s.e.m.

taken with a heparinized micropipette from an incision made in the tail during the ensuing 8 h.

Analytical method

The concentrations of tolcapone and the 3-*O*-methylated metabolite in blood were determined by a reversed-phase HPLC method (Funaki et al 1994).

Data analysis

The terminal elimination rate constant (k) was assessed by applying logarithmic regression analysis to the terminal part of the concentration–time profile. The half-life ($t_{\frac{1}{2}}$) was calculated as $t_{\frac{1}{2}} = 0{\cdot}693/k$. The area under the blood concentration (AUC)- and the first moment (AUMC)-time curves were calculated using the trapezoidal method and extrapolated to infinity. The systemic blood clearance (CL) was calculated as CL = D/AUC, in which D is the dose. The mean residence time (MRT) was calculated as MRT = AUMC/AUC. The mean residence time of the metabolite following administration of the parent drug was calculated as $MRT_m = AUMC_m/AUC_m - MRT$ (Veng-Pedersen & Gillespie 1987), where the subscript 'm'

Table 1. Pharmacokinetic parameters of oral and intravenous tolcapone administered with or without Madopar. Values are expressed as mean ± s.e.m.

Dose (mg kg^{-1})	Treatment	Compound measured	Pharmacokinetic parameter						
			C_{max} (μg mL^{-1})	T_{max} (h)	$t_{\frac{1}{2}}$ (h)	AUC (μg h mL^{-1})	MRT (h)	CL (mL min^{-1} kg^{-1})	Vd_{ss} (L kg^{-1})
Intravenous									
10	Control	Tolcapone			0·86 ± 0·09	21·8 ± 1·8	0·61 ± 0·03	7·84 ± 0·63	0·29 ± 0·04
		3-*O*-Methylated metabolite	0·37 ± 0·03	0·31 ± 0·15	2·51 ± 0·10	1·26 ± 0·05	3·11 ± 0·13		
	With Madopar	Tolcapone			0·82 ± 0·06	25·1 ± 1·2	0·70 ± 0·04	6·70 ± 0·35	0·28 ± 0·01
		3-*O*-Methylated metabolite	0·33 ± 0·03	0·50 ± 0·18	2·24 ± 0·10	1·46 ± 0·11	3·27 ± 0·23		
Oral									
20	Control	Tolcapone	15·9 ± 2·5	0·38 ± 0·06	0·81 ± 0·02	21·1 ± 1·2	1·44 ± 0·03	16·0[a] ± 1·0	
		3-*O*-Methylated metabolite	0·19 ± 0·01	0·44 ± 0·06	9·81 ± 1·88	2·50 ± 0·51	13·0 ± 3·0		
	With Madopar	Tolcapone	12·8 ± 1·0	0·75 ± 0·15	1·18 ± 0·23	24·2 ± 0·9	1·92* ± 0·08	13·9[a] ± 0·5	
		3-*O*-Methylated metabolite	0·21 ± 0·01	1·50 ± 0·84	8·75 ± 1·41	3·29 ± 0·42	11·72 ± 1·78		

* $P < 0{\cdot}05$ compared with control. [a] The value represents CL/F, in which F was 0·48 for both treatments.

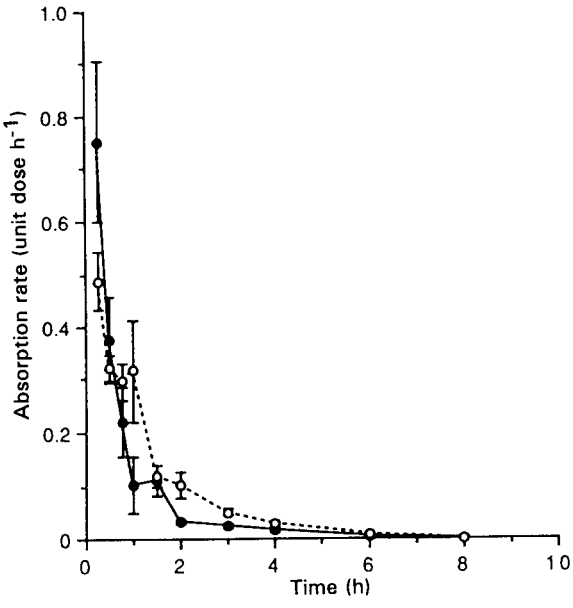

FIG. 2. Comparison of absorption rate profiles for tolcapone with Madopar (○) and without Madopar (●). Data show the mean ± s.e.m.

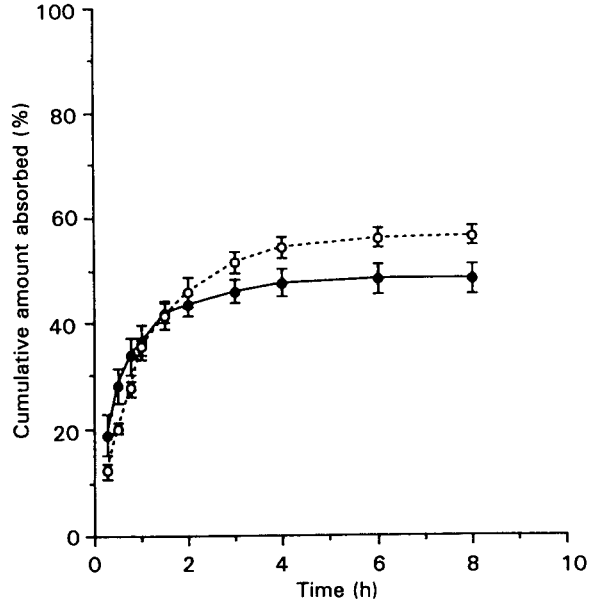

FIG. 3. Comparison of cumulative amount absorbed of tolcapone with Madopar (○) and without Madopar (●). Data show the mean ± s.e.m.

indicates metabolite. The volume of distribution at steady-state (Vd_{ss}) was calculated as $Vd_{ss} = CL \cdot MRT$. The bioavailability (F) was calculated as $F = AUC_{p.o.}/AUC_{i.v.} \cdot D_{i.v.}/D_{p.o.}$, where the subscript represents the administration route. Absorption rate and cumulative amount absorbed were calculated by a deconvolution method (Iga et al 1986), in which the characteristic response function was approximated by a biexponential function and mean characteristic response parameters in intravenous administration without Madopar were used for both treatments. The mean characteristic parameters were calculated using the nonlinear least-squares program MULTI (Yamaoka et al 1981).

Results

The blood concentration–time courses following intravenous and oral administrations of tolcapone are shown in Fig. 1. The disposition of tolcapone and its 3-*O*-methylated metabolite has been analysed previously (Funaki et al 1994).

The pharmacokinetic parameters of intravenous and oral administrations of tolcapone are shown in Table 1. There was no statistically significant difference in the pharmacokinetic parameters of tolcapone administered with or without Madopar except for the MRT in the oral administration. The absorption rate and cumulative amount absorbed are shown in Figs 2 and 3, respectively. There was no statistically significant difference in the absorption rate and cumulative amount absorbed of the drug administered with or without Madopar.

Discussion

Although the blood levels of tolcapone in the elimination phase were slightly higher when it was administered with Madopar, on both the intravenous and oral routes, there was no statistically significant difference in the pharmacokinetic parameters between treatments except for the MRT in oral administration; thus we rejected our premise that the deprivation of SAM by Madopar would change the disposition of tolcapone. Because MRT values after intravenous administration were not statistically different between treatments, the pharmacokinetics of the absorption process were investigated intensively to clarify the reason for its difference in MRT values after oral administration. However, there was no statistically significant difference in the absorption rate or cumulative amount absorbed between treatments. Although it is unclear why MRT values after oral administration were statistically different between treatments, the absorption rate of the drug with Madopar was lower than that of the drug without Madopar at the first sampling time (not significant) and it may be one of the reasons MRT values after oral administration were different. However, the lack of interaction between tolcapone and Madopar could be concluded from distribution and elimination processes as well as from the absorption process, since the difference in MRT values was small.

References

Funaki, T., Onodera, H., Ushiyama, N., Tsukamoto, Y., Tagami, C., Fukazawa, H., Kuruma, I. (1994) The disposition of the tolcapone 3-*O*-methylated metabolite is affected by the route of administration in rats. J. Pharm. Pharmacol. 46: 571–574

Iga, K., Ogawa, Y., Yashiki, T., Shimamoto, T. (1986) Estimation of drug absorption rates using a deconvolution method with nonequal sampling time. J. Pharmacokinet. Biopharm. 14: 213–225

Jeffery, D. R., Roth, J. A. (1985) Purification and kinetic mechanism of human brain soluble catechol-*O*-methyltransferase. J. Neurochem. 44: 881–885

Kaakkola, S., Gordin, A., Jarvinen, M., Wikberg, T., Schultz, E., Nissinen, E., Pentikainen, P. J., Rita, H. (1990) Effect of a novel catechol-*O*-methyltransferase inhibitor, nitecapone, on the metab-

olism of L-dopa in healthy volunteers. Clin. Neuropharmacol. 13: 436–447

Lennernäs, H., Nilsson, D., Aquilonius, S., Ahrenstedt, O., Knutson, L., Paalzow, L. K. (1993) The effect of L-leucine on the absorption of levodopa, studied by regional jejunal perfusion in man. Br. J. Clin. Pharmacol. 35: 243–250

Nissinen, E., Tuominen, R., Perhoniemi, V., Kaakkola, S. (1988) Catechol-*O*-methyltransferase activity in human and rat small intestine. Life Sci. 42: 2609–2614

Rabey, J. M., Oberman, Z., Scharf, M., Isakov, A., Bar, M., Graff, E. (1989) The influence of levodopa in the pharmacokinetics of bromocriptine in Parkinson's disease. Clin. Neuropharmacol. 12: 440–447

Rose, S., Jenner, P., Marsden, C. D. (1991) Peripheral pharmacokinetic handling and metabolism of L-dopa in the rat: the effect of route of administration and carbidopa pretreatment. J. Pharm. Pharmacol. 43: 325–330

Veng-Pedersen, P., Gillespie, W. R. (1987) A method for evaluating the mean residence time of metabolites in the body, systemic circulation, and the peripheral tissue not requiring separate i.v. administration of metabolite. Biopharm. Drug. Dispos. 8: 395–401

Yamaoka, K., Tanigawara, Y., Nakagawa, T., Uno, T. (1981) A pharmacokinetic analysis program (MULTI) for microcomputer. J. Pharmacobiodyn. 4: 879–885

Zürcher, G., Keller, H. H., Kettler, R., Borgulya, J., Bonetti, E. P., Eigenmann, R., Da Prada, M. (1990) Ro 40-7592, a novel, very potent, and orally active inhibitor of catechol-*O*-methyltransferase: a pharmacological study in rats. Adv. Neurol. 53: 497–503