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A Possible Reduction in the Renal Clearance of Ciprofloxacin by Fenbufen in Rats

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Abstract—The change in plasma concentration-time profile, serum protein binding and renal and biliary clearances of ciprofloxacin caused by coadministration of fenbufen has been studied in rats administered an intravenous dose of ciprofloxacin (5 mg kg^{-1}) alone or with fenbufen (10 mg kg^{-1}). Coadministered fenbufen significantly prolonged the plasma elimination half-life of ciprofloxacin from 40·5 to 57·6 min and tended to reduce the total body clearance of this quinolone by about 20%. The extent of ciprofloxacin binding to rat serum protein was not affected by fenbufen, nor did it affect the biliary clearance of the quinolone. However, fenbufen tended to reduce renal clearance and significantly decreased the cumulative renal excretion of the quinolone during at least the first 3 h after drug administration. These results suggest a possible reduction of ciprofloxacin clearance owing to inhibition of renal excretion by fenbufen.

The quinolonecarboxylic acids (quinolones) are new synthetic agents active against a broad spectrum of Gram-positive and Gram-negative bacteria (Neuman 1988). However, undesirable adverse reactions and drug interactions have been reported following their use. In both man (Ministry of Health & Welfare 1986, 1989) and laboratory animals (Hirai et al 1989), their concomitant administration with the non-steroidal anti-inflammatory drug, fenbufen, has given rise to serious convulsions making it essential to elucidate the mechanism of the interaction between the drugs. Previously (Katagiri et al 1989a, b; Naora et al 1990a), pharmacokinetic approaches to the possible mechanism for the convulsions induced by concomitant use of quinolones and fenbufen were described for ofloxacin, enoxacin and norfloxacin in rats. Fenbufen reduced the total body clearance of ofloxacin and prolonged the terminal elimination half-lives of both enoxacin and norfloxacin, resulting in an elevation of plasma concentrations of these quinolones in the terminal phase. The delay of quinolone elimination from plasma is likely to be related to the possible change in the renal and/or biliary excretion of the drug, as the quinolones are eliminated predominantly unchanged by renal and biliary excretion (Neuman 1988).

Since the quinolone ciprofloxacin was recently reported to induce convulsions on concomitant therapy with a non-steroidal anti-inflammatory drug (Ministry of Health & Welfare 1989), we have investigated changes in the pharmacokinetics and excretion of ciprofloxacin when coadministered with fenbufen in rats.

Materials and Methods

Chemicals

Ciprofloxacin hydrochloride was kindly supplied by Bayer AG (Leverkusen, West Germany) and fenbufen was from Lederle Japan, Ltd (Tokyo, Japan). All other reagents were commercially available and of analytical grade.

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Animals

Male Wistar rats (Clea Japan, Inc., Tokyo, Japan), 260–350 g, were lightly anaesthetized with ether and cannulated in the right jugular vein with silicone tubing (Upton 1975) about 20 h before drug administration. Animals were then kept in individual cages with free access to food and water.

Pharmacokinetic study

Ciprofloxacin hydrochloride was dissolved in distilled water and fenbufen in isotonic phosphate buffer solution containing 0·1 M sodium hydroxide (about pH 11). The rats were given a bolus dose of 5 mg kg^{-1} of ciprofloxacin alone or with 10 mg kg^{-1} of fenbufen through the jugular vein cannula. Blood samples (about 0·13 mL) were collected from the cannula, at 3, 5, 10, 20, 30, 60 and 120 min after drug administration, into heparinized tubes. Plasma was immediately separated by centrifugation.

Pharmacokinetic analysis of the plasma concentration-time data was performed assuming a two-compartment model. The non-linear least square program MULTI (Yamaoka et al 1981) was employed for regression analysis of the curves.

Serum protein binding study

Binding of ciprofloxacin to rat serum protein was determined ex-vivo, using a separate group of rats treated in the same manner as for the pharmacokinetic study. About 2·5 mL of blood was collected at 3 and 60 min after drug injection. Serum was immediately separated by centrifugation with a serum separator (Fibrichin; Takazono Sangyo Co. Ltd, Osaka, Japan) and portions (0·8 mL) of the serum were ultrafiltered using a micropartition system (MPS-3; Amicon Corp., Danvers, MA, USA) with a membrane filter at 4000 rev min^{-1} for 15 min at 37°C. The concentration of ciprofloxacin bound to serum protein was calculated by subtracting the drug concentration in the ultrafiltrate from the total concentration in serum. Non-specific adsorption of the drug to the membrane was negligible.

Clearance study

Under light ether anaesthesia, the rats with jugular vein

cannulas were also cannulated in the urinary bladder and bile duct with polyethylene tubings PE-50 and PE-10, respectively, and kept in individual restraining cages. About 60 min after the cannulation, a bolus dose of ciprofloxacin was administered alone or with fenbufen in accordance with the pharmacokinetic study. Urine and bile samples were collected during 0 to 60, 60 to 90, 90 to 120, 120 to 150 and 150 to 180 min thereafter. Immediately after the sample collection, the volume of each sample was determined. Each sample was diluted at least $\times 100$ with 0·9% NaCl (saline). A blood sample was withdrawn through the jugular vein cannula 2 min before the mid-point of each urine and bile collection period.

The renal (CL_R) and biliary (CL_B) clearances of ciprofloxacin were calculated from:

$$CL_R = U \cdot V_U/(C \cdot bw)$$
$$CL_B = B \cdot V_B/(C \cdot bw);$$

where C, U and B indicate ciprofloxacin concentrations in plasma, urine and bile, respectively; V_U and V_B indicate the flow rates of urine and bile, respectively; and bw indicates the body weight of the rat.

Analytical method

In the pharmacokinetic and serum protein binding studies, ciprofloxacin concentrations in plasma, serum and ultrafiltrate were determined by HPLC (Naora et al 1990b).

For the clearance study, a more sensitive HPLC method was developed to determine ciprofloxacin concentrations in plasma, bile and urine. A Shimadzu Model LC-4A pump (Kyoto, Japan) equipped with a Shimadzu Model RF-530 fluorescence spectromonitor (excitation, 277 nm: emission, 445 nm) was used with a stationary phase of Chemcosorb 5-ODS-H (Chemco Scientific Co. Ltd, Osaka, Japan) packed in a stainless steel column (4·6 mm i.d. × 150 mm). The mobile phase was methanol–0·01 M monopotassium phosphate (3:2, pH 2·5) containing 2 mM sodium lauryl sulphate. The flow rate and column temperature were maintained at 0·8 mL min^{-1} and 40°C, respectively. Ciprofloxacin was extracted from plasma, diluted urine and bile samples according to Naora et al (1990b) before chromatographic analysis.

Results

Plasma concentration-time profile

The average plasma concentration-time curves of ciprofloxacin after bolus intravenous administration with or without fenbufen are shown in Fig. 1. In both groups, the plasma concentration of ciprofloxacin was found to decline biexponentially with time. A significant elevation of the plasma concentration in the terminal phase occurred after the coadministration with fenbufen. Pharmacokinetic parameters for ciprofloxacin based on the plasma concentration-time data of individual rats are summarized in Table 1. The elimination half-life at the terminal phase $t_{\frac{1}{2}}\beta$ was 40·5 min in rats given CPFX alone and was significantly prolonged to 57·6 min by the coadministration of fenbufen. Coadministered fenbufen tended to reduce the total body clearance (CL) and to increase the area under the plasma concentration-time curve (AUC) of ciprofloxacin.

Table 1. Pharmacokinetic parameters for ciprofloxacin after bolus intravenous administration with or without fenbufen in rats.

	Parameter	Alone	With fenbufen
A	($\mu g\ mL^{-1}$)	9·02±4·44	9·40±12·9
α	($10^{-2}\ min^{-1}$)	38·3±11·6	30·7±19·3
B	($\mu g\ mL^{-1}$)	1·59±0·22	1·44±0·06
β	($10^{-2}\ min^{-1}$)	1·74±0·25	1·23±0·20*
$t_{\frac{1}{2}}\beta$	(min)	40·5±6·1	57·6±10·7*
AUC	($\mu g\ min\ mL^{-1}$)	115·7±20·5	142·8±18·3
CL	($mL\ min^{-1}\ kg^{-1}$)	44·3±7·7	35·5±4·7
V_c	($mL\ kg^{-1}$)	532±179	836±419
V_p	($mL\ kg^{-1}$)	1555±215	1643±158

Each parameter is estimated from the plasma concentration (C)-time (t) data by fitting to the two-compartment model ($C = Ae^{-\alpha t} + Be^{-\beta t}$, where A, B, α and β are hybrid parameters). Each value represents the mean ± s.d. of five rats. V_c and V_p are the distribution volumes of central and peripheral compartments, respectively.
*$P < 0.05$ compared with ciprofloxacin alone.

Serum protein binding

The results of the protein binding study are summarized in Table 2. At 3 and 60 min after drug administration, the total (unbound + bound) concentrations of ciprofloxacin were 2·63 and 0·501 $\mu g\ mL^{-1}$, respectively, and the fractions bound to serum protein were 25·0 and 22·2%, respectively. Binding was not affected by coadministration of fenbufen.

Renal and biliary excretion

Fig. 2 shows the plasma concentrations of ciprofloxacin at three intervals for urine and bile collection (90 to 180 min after drug administration). Plasma concentrations of ciprofloxacin tended to increase on coadministration with fenbufen, as with the pharmacokinetic study. The clearance value in individual rats was estimated as the average of the three values obtained from these intervals.

The CL_R and CL_B of ciprofloxacin are shown in Fig. 3. In

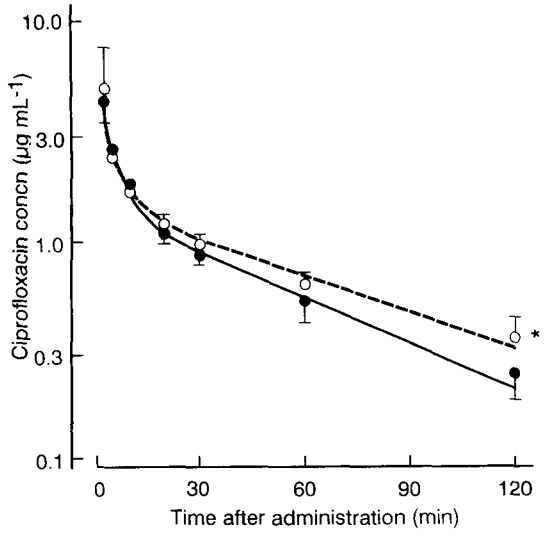

FIG. 1. Plasma concentration-time profiles of ciprofloxacin after bolus intravenous administration (5 mg kg^{-1}) with (○) or without (●) fenbufen (10 mg kg^{-1}) in rats. Each point and vertical bar indicate the mean and s.d. of five rats. The lines represent the computer-fitted biexponential curves for the mean data [Weight(i) = 1/Ci, where C is the plasma drug concentration]. *$P < 0.05$ compared with ciprofloxacin alone.

Table 2. Serum protein binding of ciprofloxacin after bolus intravenous administration with or without fenbufen in rats.

Time (min)	Treatment	Serum drug concn (μg mL^{-1}) Total	Unbound	Fraction bound (%)
3	Alone	2·63 ± 0·10	1·97 ± 0·14	25·0 ± 3·7
	With fenbufen	3·11 ± 0·96	2·37 ± 0·63	22·9 ± 4·1
60	Alone	0·501 ± 0·066	0·387 ± 0·033	22·2 ± 5·9
	With fenbufen	0·565 ± 0·060	0·425 ± 0·045	24·7 ± 5·4

T, time after drug administration. Each value represents the mean ± s.d. of five rats.

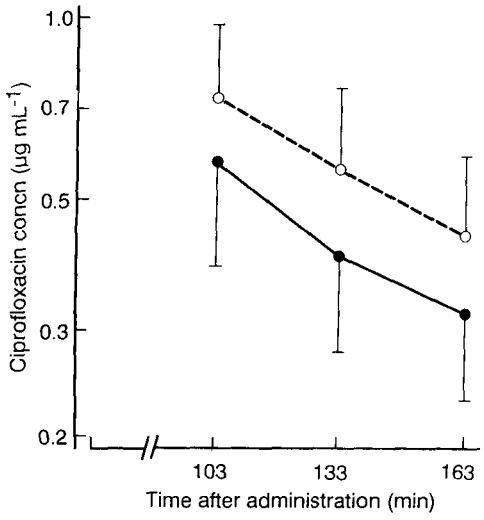

FIG. 2. Plasma concentrations of ciprofloxacin after intravenous administration with (○) or without (●) fenbufen in the clearance study. Each point and vertical bar indicate the mean and s.d. of five rats.

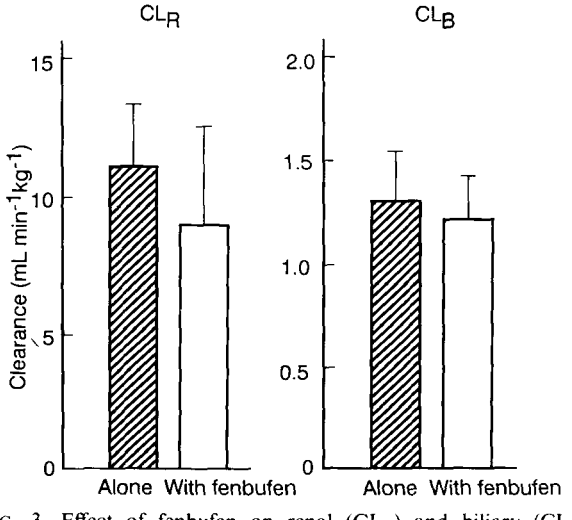

FIG. 3. Effect of fenbufen on renal (CL_R) and biliary (CL_B) clearances of ciprofloxacin. Each column and vertical bar indicate the mean and s.d. of five rats.

rats given ciprofloxacin alone, the CL_R was 11·1 ± 2·2 mL min^{-1} kg^{-1}. In the coadministered group, CL_R was 9·06 ± 3·45 mL min^{-1} kg^{-1}. No change was observed in the CL_B of ciprofloxacin on coadministration of fenbufen.

Cumulative renal and biliary excretion time-courses of

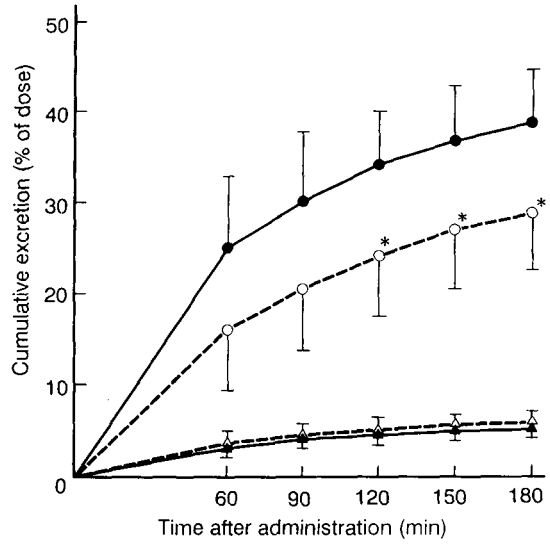

FIG. 4. Cumulative excretion of ciprofloxacin in urine and bile after intravenous administration with (○ urine, △ bile) or without (● urine, ▲ bile) fenbufen. Each point and vertical bar indicate the mean and s.d. of five rats. *$P < 0.05$ compared with ciprofloxacin alone.

ciprofloxacin are shown in Fig. 4. The cumulative renal excretion of ciprofloxacin decreased on coadministration of fenbufen and significant differences were observed during 120 to 180 min after drug administration. No change was observed in the amount of ciprofloxacin excreted into bile.

Discussion

The results (Fig. 1, Table 1) show the terminal elimination half-life of ciprofloxacin to be significantly prolonged about 1·5 times by the coadministration of fenbufen, and the total body clearance of ciprofloxacin tending to be reduced by about 20%. Since fenbufen and its active metabolite, 4-biphenylacetic acid (felbinac), are known to bind strongly to human serum protein (Chiccarelli et al 1980), the displacement of ciprofloxacin from its binding site by fenbufen and/or felbinac may be the cause of this change in pharmacokinetics. However, ciprofloxacin binding to serum protein was unchanged by the coadministration of fenbufen (Table 2), suggesting that the elevation of plasma ciprofloxacin concentration observed in the terminal phase is not related to its binding to serum protein.

Ciprofloxacin, like other quinolones, is predominantly excreted unchanged in urine (Siefert et al 1986). It has also been reported that ciprofloxacin is excreted in bile in man (Tanimura et al 1986). In the present study, however, the contribution of biliary excretion to the elimination of ciprofloxacin from the blood was much lower than that of renal excretion. As shown in Fig. 3, coadministered fenbufen tended to reduce the renal clearance of ciprofloxacin by about 20%, and the cumulative amount of ciprofloxacin excreted in urine was significantly decreased by the coadministration of fenbufen (Fig. 4). These results indicate that fenbufen may inhibit its renal excretion which is a major elimination route of ciprofloxacin. Gasser et al (1987) reported that the reduction in renal clearance in patients with

impaired renal function changed the elimination half-life, total body clearance and area under serum concentration-time curve of ciprofloxacin. An active tubular secretion mechanism is likely to be involved in the elimination of ciprofloxacin, since its renal clearance is greater than normal creatinine clearances (Höffler et al 1984; Wingender et al 1984b). Wingender et al (1984a) demonstrated that the plasma elimination half-life of ciprofloxacin was prolonged by the administration of probenecid, an inhibitor of active secretion processes due to its high affinity for anion carriers. Generally, organic acids are secreted at renal proximal tubules by an anion transport system (Møller & Sheikh 1982). Chiba et al (1985, 1986) have reported that the elimination of sulfamethizole, a sulphonamide known to be predominantly excreted by renal tubular secretion via an anion transport mechanism, is prolonged by the coadministration of fenbufen or felbinac. It is suggested, therefore, that the slight reduction in the renal clearance of ciprofloxacin by the coadministration of fenbufen may be due to competitive inhibition for renal tubular secretion by an anion transport mechanism.

The plasma concentration-time profiles and resultant pharmacokinetic parameters for both fenbufen and felbinac were not affected by the coadministration of ciprofloxacin (unpublished data). These results are similar to those obtained in our previous studies for other quinolones (Katagiri et al 1989a, b; Naora et al 1990a).

It is proposed that the elevation in plasma concentration of ciprofloxacin by administered fenbufen may raise the concentration of the quinolone in the central nervous system (CNS), leading to the induction of neurotoxic side-effects, including convulsions, as a result of the pharmacodynamic interaction, i.e. enhancement of the inhibitory effect of the quinolone on γ-aminobutyric acid binding to its receptors, as proposed by Tsuji et al (1988a, b). The slight alteration of ciprofloxacin pharmacokinetics by coadministered fenbufen may be a possible but not the only reason for the observed side effects in the CNS.

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