Journal of Chromatography, 183 (1980) 246—249
Biomedical Applications
Elsevier Scientific Publishing Company, Amsterdam — Printed in The Netherlands

CHROMBIO. 590

Note

Determination of carpipramine in plasma by high-performance liquid chromatography

TSUNEO SADANAGA, KOZO HIKIDA, KATSUNOBU TAMETO and MICHIO NAKANISHI

Research Laboratories, Yoshitomi Pharmaceutical Industries Ltd., Yoshitomi-cho, Fukuoka 871 (Japan)

and

YOSHIKAZU MATSUSHIMA and YOSUKE OHKURA*

Faculty of Pharmaceutical Sciences, Kyushu University 62, Maidashi, Higashi-ku, Fukuoka 812 (Japan)

(Received February 5th, 1980)

Carpipramine, 1′-[3-(10,11-dihydro-5H-dibenz[*b*,*f*]azepin-5-yl)-propyl]-[1,4′-bipiperidine]-4′-carboxamide (CPP) (Fig. 1), is a currently used psychotropic agent [1]. CPP is usually administered orally and is absorbed rapidly from the gastrointestinal tract. Several preparations containing its dihydrochloride monohydrate have been used clinically.

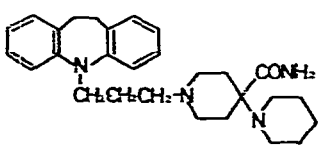


Fig. 1. Structure of carpipramine.

The pharmacological characteristics of CPP [2] and its metabolic fate in rats and rabbits [3, 4] have been reported. In these early studies, CPP was determined spectrophotometrically or fluorimetrically in extracts from tissue homogenates and body fluids [5].

For a comparison of the bioavailabilities of CPP and its preparations and for

0378-4347/80/0000-0000/$02.25

drug monitoring during therapy, a more specific and sensitive method of assay was required. High-performance liquid chromatography (HPLC) was found to meet this demand. In this paper, we describe the HPLC determination of CPP in the plasma of dogs, though the method can be applied to human plasma and tissue homogenates.

EXPERIMENTAL

Materials

Carpipramine dihydrochloride monohydrate, and its various preparations, and chlorpromazine hydrochloride were the products of Yoshitomi Pharmaceutical Industries Ltd., Fukuoka, Japan. All solvents and chemicals were of reagent grade.

HPLC instrumentation

An Hitachi Model 635 liquid chromatograph equipped with a universal injector and an Hitachi variable-wavelength UV effluent monitor operated at 250 nm was used. The column was a Zorbax-SIL (Du Pont, Wilmington, DE, U.S.A.; particle size 5 μm; 150 mm × 3.9 mm I.D.). The flow-rate of the mobile phase was 0.4 ml/min.

Extraction procedure

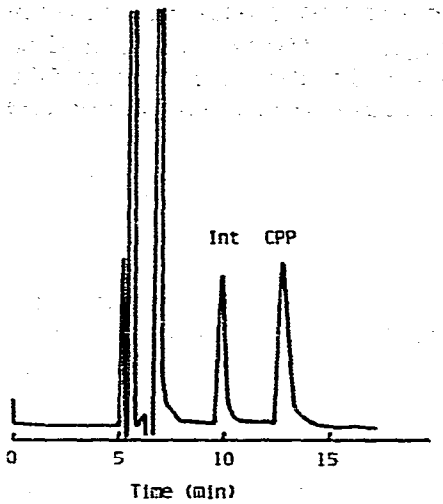
Plasma (0.3—3.0 ml), 10% NaOH (1.0 ml), water (2.0 ml), and *n*-heptane containing 1.5% isoamyl alcohol (25 ml) were added to a 50-ml test-tube with a ground-glass stopper. The amount of plasma was varied according to the CPP content and available volume. The sample was extracted for 20 min with vigorous shaking. The organic layer (20 ml) was separated by centrifugation and shaken for 10 min with 10 *M* HCl (5.0 ml).

After the heptane layer was discarded, the aqueous HCl layer (4.5 ml) was transferred to a glass-stoppered test-tube. The aqueous solution (1.0 ml) containing chlorpromazine hydrochloride (200 ng/ml) as an internal standard and 40% NaOH (0.5 ml) were added. The resulting solution was extracted with chloroform (100 μl) for 10 min with vigorous shaking. After the removal of the aqueous layer, a 30-μl volume of the chloroform solution was injected into the liquid chromatograph.

For the preparation of the calibration curve, CPP was dissolved in water so as to contain 100—700 ng/ml. Drug-free plasma (3.0 ml), CPP solution (2.0 ml), and 10% NaOH (1.0 ml) were mixed. These standards were carried through the procedure described above.

RESULTS AND DISCUSSION

Fig. 2 shows a chromatogram of an extract of plasma of a dog administered 100 mg of CPP orally. The mobile phase used was dichloromethane containing 10% methanol and 0.2% aqueous ammonia. The retention times were 12.6 min for CPP and 9.7 min for chlorpromazine, the internal standard. The ratio of the peak height of CPP to that of the internal standard was plotted against the



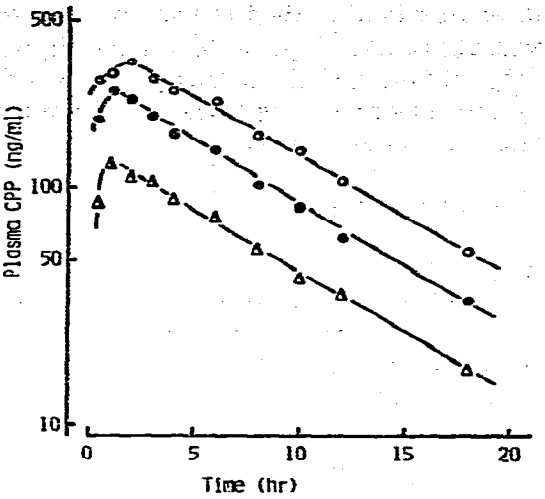


Fig. 2. Chromatogram of an extract of plasma of a dog administered 100 mg of CPP orally. Int = chlorpromazine.

Fig. 3. Plasma concentration of CPP at various times after oral administration to dogs. Doses of CPP per animal were 150 mg (○), 100 mg (●), and 50 mg (△). The analytical data presented are the averages of three animals.

amounts of CPP added to the standard. The calibration curves thus obtained were linear up to at least 700 ng and passed through the origin.

It was verified that the extraction of CPP and chlorpromazine into chloroform from aqueous alkaline solution were virtually complete in their wide ranges of concentration. In the determination of CPP in plasma containing 100 ng/ml, the standard deviation was 3 ng (n = 10; the volume of plasma used, 3 ml).

The present method permits the accurate determination of CPP in plasma at concentrations as low as 9 ng/ml (the volume of plasma, 3 ml) and is suited for monitoring the drug in the therapeutic dose range (50—300 mg/day per person).

Fig. 3 shows examples of the time—concentration curves in the plasma of dogs administered CPP. The CPP concentration in plasma increased immediately after the oral administration, reached a maximum in 1—2 h, and then decreased at a first-order rate. The biological half-life was about 6.0 h. These measurements give the basis for comparing the bioavailabilities of CPP and its various preparations. The details of the pharmacokinetics will be discussed elsewhere.

Since the present assay method was introduced in our laboratories, unfailing analytical results have always been obtained.

REFERENCES

1 M. Nakanishi and T. Munakata, Jap. Pat., 488776 (1967).
2 M. Nakanishi, T. Okada and T. Tsumagari, Arzneim.-Forsch., 18 (1968) 1435.

3 M. Nakanishi, Y. Kato, M. Setoguchi, A. Tsuda and H. Yasuda, Yakugaku Zasshi, 90 (1970) 197.
4 M. Nakanishi H. Imamura, E. Matsui and Y. Kato, Yakugaku Zasshi, 90 (1970) 204.
5 M. Nakanishi, Y. Kato and H. Yasuda, Yakugaku Zasshi, 90 (1970) 193.