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Determination of sultopride and tiapride in serum by gas chromatography using a surface ionisation detector

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ABSTRACT

A sensitive and selective method has been developed for the determination of sultopride and tiapride in serum using gas chromatography with a surface ionisation detector. No interfering peaks from endogenous substances were observed. The method showed good reproducibility and accuracy, and the standard curve was linear up to 2 μg/ml with a correlation coefficient of 0.999. This method is applicable to pharmacokinetic studies and therapeutic drug monitoring of sultopride and tiapride.

INTRODUCTION

Sultopride, N-[(1-ethyl-2-pyrrolidinyl)methyl]-5-(ethylsulphonyl)-2-methoxybenzamide, and tiapride, N-[2-diethylamino)ethyl]-5-(methylsulphonyl)-2-methoxybenzamide (Fig. 1) are antagonists of dopaminergic receptors, structurally classified as substituted benzamides [1]. Both drugs, in addition to sulpiride, selectively inhibit D_2 dopaminergic receptors and are distinct from typical neuroleptics such as phenothiazines and butyrophenones [2]. Their pharmacological and clinical profiles are considered to be different from each other; sultopride is used as an antipsychotic agent, whereas tiapride is used to ameliorate cerebral circulation and metabolism.

Little information is available on the pharmacokinetic behaviours of these drugs in humans. Several analytical methods have been described employing

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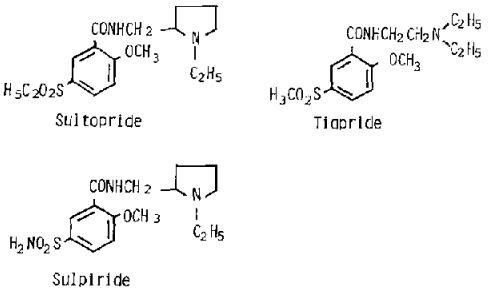

Fig. 1. Structures of sultopride, tiapride and sulpiride.

high-performance liquid chromatography (HPLC) [3–5], radioimmunoassay (RIA) [6], and enzyme immunoassay [7], but no gas chromatographic (GC) method has yet been proposed. A surface ionisation detector (SID), a new detector for GC, shows a sensitive and selective response to amines, especially to aliphatic tertiary amines [8,9].

This paper presents a GC method for the determination of sultopride and tiapride using a SID.

EXPERIMENTAL

Materials

Sultopride hydrochloride and tiapride hydrochloride were kindly supplied by Mitsui Pharmaceuticals (Tokyo, Japan) and Fujisawa Pharmaceutical (Osaka, Japan), respectively. Stock solutions were prepared using distilled water and were stored at 4°C. Chloroform was distilled before use. All other reagents were of analytical-reagent grade and were purchased from Wako Pure Chemical Industries (Osaka, Japan).

Extraction procedure for sultopride

To a 1.0-ml portion of serum in a 10-ml glass test-tube, 10 μl of a tiapride solution as an internal standard (5 μg/ml), 500 μl of 0.5 *M* sodium hydrochloride solution saturated with sodium chloride and 3 ml of chloroform were added. The mixture was agitated for 10 min by a reciprocal shaker and centrifuged for 5 min at 1000 *g*. The organic phase was then transferred into another test-tube and evaporated to dryness using a rotary vacuum evaporator. The residue was dissolved in 50 μl of chloroform and 4-μl aliquots of the resulting solution were injected into the GC system.

Extraction procedure for tiapride

Tiapride was extracted from serum in a similar manner to sultopride. A 10-μl volume of sultopride solution (10 μg/ml) was added as an internal standard. The

residue was dissolved in 0.3–1 ml of chloroform and 4-μl aliquots of the resulting solution were injected into the GC system.

Instruments

A GC–RIA system (Shimadzu, Kyoto, Japan) was used. The system was equipped with a flexible fused-silica capillary column (DB-5, 30 m × 0.24 mm I.D., film thickness 1.0 μm, J&W Scientific, Folsom, CA, U.S.A.). A moving-needle solventless sample injector was used.

The structural design of the SID (Shimadzu) is shown in Fig. 2. The SID is a selective detector for GC which uses an electrically heated platinum filament as an emitter surface. The detection mechanism is considered to be the positive surface ionisation of chemical species on the hot surface [8,9].

Helium was used as a carrier and make-up gas. The operating temperatures were: injection port, 310°C; column, 300°C.

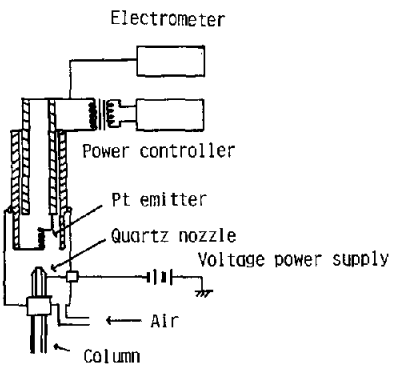

Fig. 2. Schematic diagram of the surface ionisation detector.

Standard curves

Standard curves were obtained by adding an internal standard and known amounts of sultopride or tiapride (0.05–2 μg/ml) in distilled water to a blank serum sample. Extraction was carried out under the described experimental conditions. The concentrations of both drugs were estimated by the peak-area ratios of both drugs to the internal standard. Standard curves were obtained for each set of serum samples.

Serum samples were obtained from a healthy male volunteer receiving 200 mg of sultopride after obtaining informed consent.

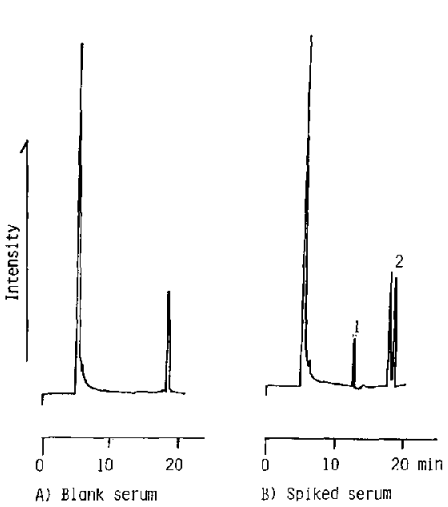

Fig. 3. Chromatograms of (A) a blank serum extract and (B) an extract of serum spiked with tiapride (50 ng) and sultopride (500 ng). Peaks: 1 = tiapride; 2 = sultopride.

RESULTS

Gas chromatography

Typical chromatograms obtained from the analysis of blank serum and serum spiked with sultopride and tiapride are shown in Fig. 3. The retention times were 13.1 and 18.5 min for sultopride and tiapride, respectively. As shown in Table I, a good separation of sultopride and tiapride from other antipsychotic and anticholinergic agents was observed. The whole procedure to determine sultopride and tiapride in patient serum was completed within 2 h.

TABLE I

RETENTION TIMES OF SULTOPRIDE, TIAPRIDE AND OTHER ANTIPSYCHOTIC AND ANTICHOLINERGIC AGENTS

Drug	Retention time (min)
Trihexyphenizyl	7.0
Biperiden	7.9
Chlorpromazine	9.6
Levopromazine	9.7
Zotepine	10.2
Tiapride	13.1
Haloperidol	16.4
Sultopride	18.5

TABLE II

RECOVERY VALUES OF SULTOPRIDE AND TIAPRIDE AFTER EXTRACTION FROM SERUM

Drug	Concentration (μg/ml)	Recovery (mean ± S.E.M., $n = 6$) (%)
Sultopride	0.5	93.3 ± 1.0
	1.0	94.0 ± 0.7
	2.0	92.3 ± 2.3
Tiapride	0.5	98.0 ± 1.3
	1.0	100.5 ± 1.8

Validation of the method

The minimum detectable limits for sultopride and tiapride were 4 ng and 200 pg, respectively. The standard curves were linear over the range 0.05–2 μg/ml, with a correlation coefficient of 0.999 for both drugs.

Table II summarises the results of recovery experiments from spiked sera. The mean extraction efficiency of both drugs in spiked sera was at least 90%. No effort was made to increase the recovery value of either agent. The within- and between-day reproducibility of the method was checked for five serum concentrations. The within- and between-day coefficients of variation for sultopride were 0.2–2.0 and 0.4–17%, respectively, at serum concentrations of 0.1–2.0 μg/ml (Tables III and IV). Those for tiapride were calculated to be 0.0–4.0% and 0.5–6.0%, respectively, at serum concentrations of 0.05–1 μg/ml (Tables III and IV).

TABLE III

WITHIN-DAY COEFFICIENTS OF VARIATION IN DETERMINATION OF SULTOPRIDE AND TIAPRIDE

Drug	Concentration (μg/ml)		C.V. (%)
	Added	Found (mean ± S.E.M., $n = 6$)	
Sultopride	0.1	0.106 ± 0.007	1.05
	0.5	0.482 ± 0.004	2.04
	1.0	0.962 ± 0.006	0.54
	2.0	2.02 ± 0.009	1.32
Tiapride	0.05	0.063 ± 0.003	3.96
	0.1	0.100 ± 0.002	0.55
	0.2	0.200 ± 0.004	0.76
	0.5	0.480 ± 0.007	1.96
	1.0	1.01 ± 0.003	0.00

TABLE IV

BETWEEN-DAY COEFFICIENTS OF VARIATION IN DETERMINATION OF SULTOPRIDE AND TIAPRIDE

Drug	Concentration (μg/ml)		C.V. (%)
	Added	Found (mean ± S.E.M., $n = 6$)	
Sultopride	0.1	0.128 ± 0.022	17.2
	0.5	0.480 ± 0.002	0.31
	1.0	0.940 ± 0.022	2.39
	2.0	1.98 ± 0.020	2.02
Tiapride	0.05	0.060 ± 0.004	6.04
	0.1	0.099 ± 0.001	0.76
	0.2	0.200 ± 0.001	0.25
	0.5	0.480 ± 0.070	1.45
	1.0	1.00 ± 0.005	0.50

The developed method was applied to determine the concentrations of sultopride in serum from a healthy volunteer after oral administration of 200 mg of sultopride. No peaks other than those of the sultopride and tiapride (internal standards) were observed in the GC profiles of the serum extracts. A concentration–time curve of sultopride after the oral administration of sultopride is shown in Fig. 4. The disposition pattern was fitted to the two-compartment open model with intravenous injection because the maximum concentration was observed at the first sampling point of 0.5 h. The calculated pharmacokinetic parameters of sultopride are shown in Table V.

TABLE V

PHARMACOKINETIC PARAMETERS OF SULTOPRIDE IN A HEALTHY MALE VOLUNTEER RECEIVING 200 mg OF SULTOPRIDE

Parameter[a]	Value
V_1/f (l)	50.4
k_{12} (h^{-1})	0.347
k_{21} (h^{-1})	0.302
k_{el} (h^{-1})	0.360
AUC (μg h ml^{-1})	8.32
MRT (h)	2.37

[a] Abbreviations: (V_1) volume of distribution of the central compartment; (f) fraction absorbed; (k_{12}) transfer rate constant from the central to the peripheral compartment; (k_{21}) transfer rate constant from the peripheral to the central compartment; (k_{el}) elimination rate constant from the central compartment; (AUC) area under the curve; (MRT) mean residence time.

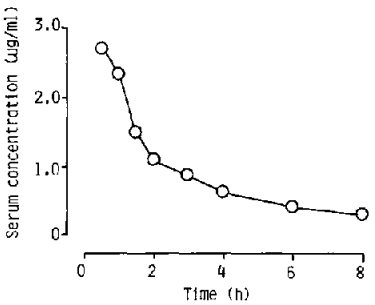

Fig. 4. Concentration–time curve of sultopride in serum after oral administration of 200 mg of sultopride to a healthy male volunteer.

DISCUSSION

Several analytical methods for the determination sultopride and tiapride have been described previously, mostly employing HPLC [3–5]. However, these methods seem to be less selective than the analytical method reported here using capillary GC with SID.

The SID has been reported to show an extremely high response to aliphatic tertiary amimes [8,9]. Sulpiride and sultopride are classified as benzamide derivatives with a tertiary amino group. The response of sultopride to SID was several times higher than that of sulpiride. Tiapride showed a superior response to sultopride and sulpiride, possibly because the tertiary amino group of tiapride is not part of a ring structure. Sultopride and tiapride were determined in serum with a good sensitivity, but the sensitivity of the method in sulpiride was relatively poor.

Substituted benzamides are rapidly becoming important psychotherapeutic agents. Recently, it was reported for sulpiride that differences in pharmacokinetic parameters were indicated between healthy volunteers and patients with renal function impairment [10]. In spite of this, few pharmacokinetic and clinical studies have so far been carried out on sultopride and tiapride in disease states [9]. This GC method using SID will be applicable to the pharmacokinetic study of benzamides.

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