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Note

Determination of therapeutic levels of butriptyline in plasma by gas–liquid chromatography

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Depressive illness is most often treated with a group of drugs known as the tricyclic antidepressants[1]. The number of these drugs has steadily increased since Kuhn first introduced imipramine[2,3] and a recent addition is butriptyline (*d,l*-10,11-dihydro-β,N,N-trimethyl-5H-dibenzo[*a,d*]cycloheptene-5-propylamine hydrochloride; see Fig. 1). Clinical trials have shown that this compound is effective in the treatment of depressive illness[4].

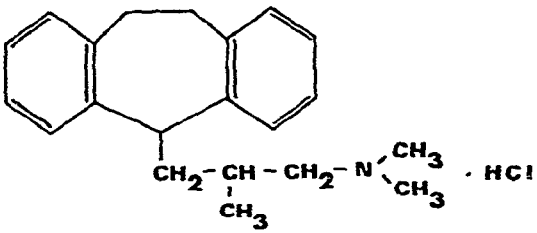

Fig. 1. Structure of butriptyline.

However, there is little information on the plasma levels achieved by the patients in these trials, due to the lack of a sensitive analytical method.

In this paper a gas–liquid chromatographic (GLC) method is described which allows determination of therapeutic levels of butriptyline in plasma. The method is suitable for routine application and for the assessment of pharmacokinetic data.

EXPERIMENTAL

Chemicals and reagents

n-Hexane (Nanograde; Mallinckrodt) was redistilled after washing with an aqueous, acidified solution of potassium permanganate and dried over anhydrous sodium sulphate. All other chemicals were of analytical reagent grade (Univar, Ajax, Sydney, Australia).

Butriptyline hydrochloride was supplied by Ayerst (Sydney, Australia) and maprotiline hydrochloride was supplied by Ciba-Geigy (Sydney, Australia). Both compounds were used as received.

Extraction procedure

Samples of 4 ml of plasma were placed in a 20-ml glass test tube, followed by 0.3 ml of a 4-μg/ml solution of maprotiline hydrochloride in ethanol as internal standard. The solution was alkalinised (pH >11–12) with 0.5 ml of 5 *M* sodium hydroxide and extracted with 5 ml of *n*-hexane by shaking for 20 min. The tube was centrifuged at 1400 *g* for 5 min and the hexane phase separated. A further 5-ml portion of *n*-hexane was added to the plasma and the extraction repeated. The two hexane extracts were combined and shaken with 1 ml of 1 *M* hydrochloric acid for 15 min and the tube centrifuged at 1400 *g* for 5 min. The organic phase was discarded and the aqueous phase was transferred to a 5-ml test tube and made alkaline with 0.3 ml of 5 *M* sodium hydroxide. The free base was extracted by shaking the solution with 1 ml of *n*-hexane for 10 min and then centrifuging at 1400 *g* for 5 min. This hexane phase was separated and the remaining solution further extracted with another 1 ml of hexane. The hexane extracts were combined, 0.5 ml of a 0.04% solution of triethanolamine in chloroform added and the solution evaporated to dryness at 35–40° in a stream of nitrogen. The solid so obtained was reconstituted in 40 μl of hexane and 5 μl samples injected into the gas chromatograph.

Gas chromatography

The analysis was carried out on a Hewlett-Packard Model 419 gas chromatograph equipped with a flame ionisation detector with the following conditions: column, coiled glass 1.8 m × 4 mm I.D.; liquid phase, 3% OV-17 on Supelcoport 80–100 mesh (Supelco); column temperature, 215°; detector and injection port temperature, 260°; carrier gas (nitrogen) flow-rate, 21 ml/min. Retention times and peak areas were determined with a Hewlett-Packard Model 3380A reporting integrator.

Calibration graphs

A stock solution of butriptyline in ethanol containing 1 mg/ml, was prepared and used for the preparation of plasma standard solutions containing 0, 50, 100, 200 and 400 ng of butriptyline per ml. These plasma standards were then carried through the entire extraction procedure as described and the ratio of peak area of butriptyline to peak area of maprotiline calculated. This ratio was then plotted against butriptyline concentration and linear regression analysis gave a straight line with a correlation coefficient greater than 0.99 (Fig. 2). The calibration graph was used to determine the levels achieved by patients on therapeutic doses.

RESULTS AND DISCUSSION

The retention times observed for butriptyline, maprotiline and some other tricyclic antidepressant drugs are shown in Table I. Extraction of blank plasma showed slight responses corresponding with the retention time of butriptyline, equivalent to 5 ng/ml of the drug. Thus the limit of detection for butriptyline was considered to be 10 ng/ml. No response was detected at the retention time corresponding to maprotiline (see Fig. 3). A small unidentified peak appeared at a retention time between that of butriptyline and maprotiline. This peak possibly arises from an impurity in the solvents used and does not interfere with the analysis, as shown by the linearity of the calibration graph.

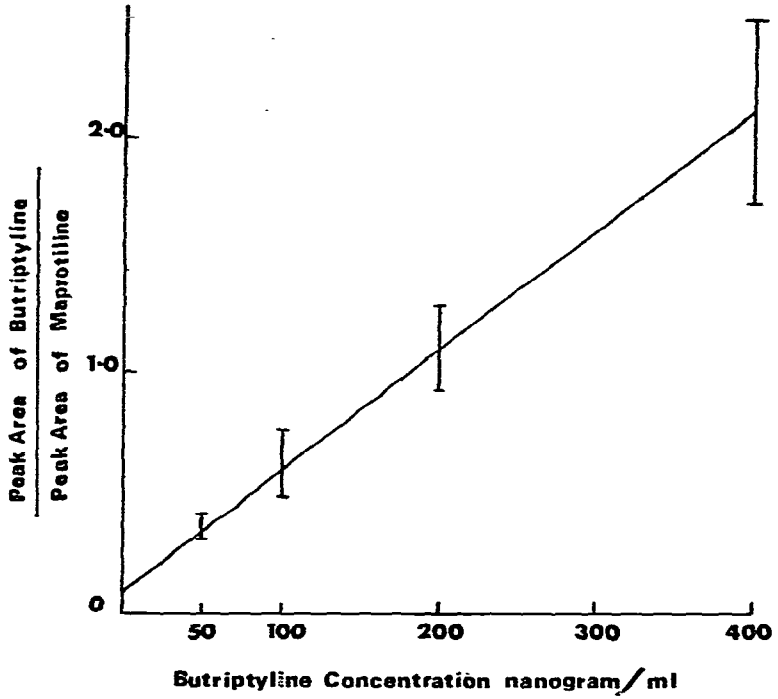

Fig. 2. Standard curve for butriptyline determination. The curve obtained is the average of four determinations, values are the mean ± standard deviation. Regression equation obtained: Peak area ratio = 0.005 (butriptyline concentration) + 0.096. Correlation coefficient > 0.99.

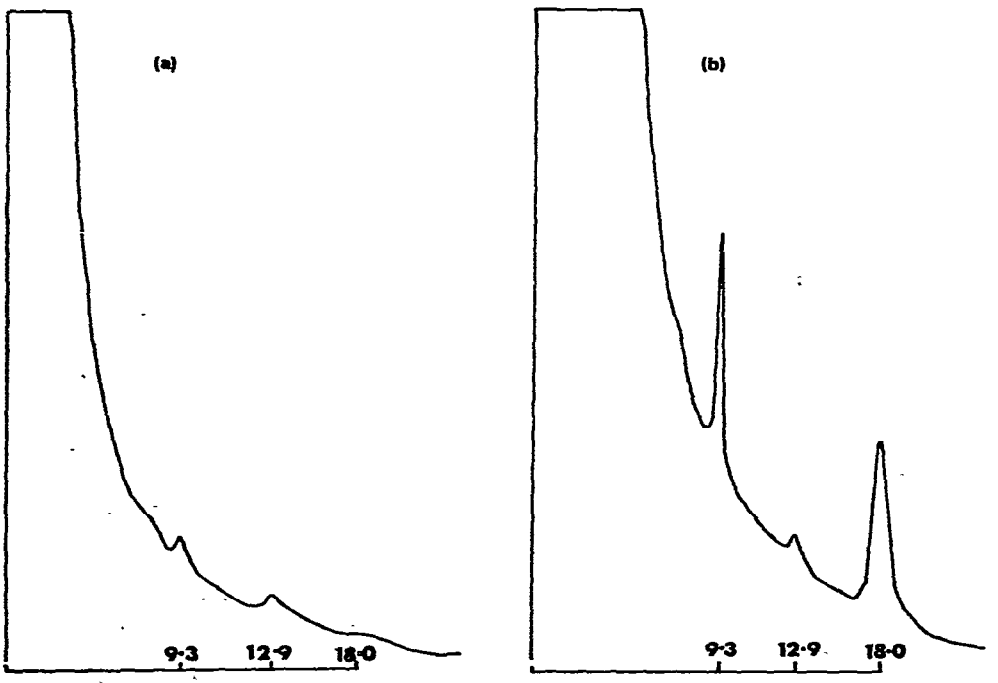

Fig. 3. Gas–liquid chromatograms of (a) blank plasma and (b) an extracted plasma standard containing 100 ng/ml of butriptyline and 300 ng/ml of maprotiline. The small peak at 12.9 min is an impurity arising from the solvents.

The analytical recovery was 65–70% for butriptyline, determined by comparison of relative peak areas of extracted plasma samples with those of non-extracted ethanol standards. This incomplete recovery is corrected for by the use of an internal standard (maprotiline) and by use of extracted plasma standards in the construction of the calibration graph.

TABLE I

GLC RETENTION TIMES OF BUTRIPTYLINE, MAPROTILINE AND OTHER TRICYCLICS

Conditions, see Experimental.

Compound	*Retention time (min)*
Butriptyline	9.3
Amitriptyline	9.7
Nortriptyline	10.8
Doxepin	11.2
Desmethyldoxepin	13.0
Protriptyline	13.0
Maprotiline	18.0

Plasma concentrations of butriptyline were determined on different days from four patients each receiving 150 mg/day. The results are listed in Table II and range from 50 to 205 ng/ml, values well within the sensitivity of the method. No other metabolites of butriptyline were detected in the chromatogram of the extracted plasma. Considering the structure of butriptyline and the usual metabolic pathways of tricyclics[5], a number of metabolites would be feasible *viz.*, N-demethyl-, N-oxide-,

TABLE II

PLASMA CONCENTRATIONS OF BUTRIPTYLINE (ng/ml) OBTAINED FROM PATIENTS RECEIVING 150 mg/day

Patient	*Day 7*	*Day 14*	*Day 21*
J.L.	180	205	185
K.W.	145	165	145
B.P.	130	50	—
G.M.	150	102	90

2-hydroxy- or 10-hydroxybutriptyline. Of these N-demethyl-butriptyline is the most likely metabolite. The absence of any of these metabolites in the chromatogram could be due to loss or non-extraction during the procedure, levels so low that they are not detectable, or a retention time within the solvent, butriptyline or maprotiline peaks. In the absence of any pure demethylated compound these possibilities have not been investigated. However, based on the chromatographic behaviour of other demethylated metabolites of tricyclic antidepressants (see Table I), undetectable levels appears to be the most likely explanation.

The method described here allows the determination of steady-state plasma levels of butriptyline in patients receiving therapeutic doses of the drug. It is suitable for pharmacokinetic and toxicological estimations.

REFERENCES

1 G. D. Burrows, *Drugs*, 11 (1976) 209.
2 R. Kuhn, *Schweiz. Med. Wochenschr.*, 87 (1957) 1135.
3 R. Kuhn, *Amer. J. Psychiat.*, 115 (1958) 459.
4 H. Grivois, *J. Med.*, 2 (1971) 276.
5 L. F. Gram, *Dan. Med. Bull.*, 21 (1974) 218.