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Letter to the Editor

Determination of 5-hydroxypropafenone in biological fluids by fused-silica capillary gas chromatography using electron-capture detection

Sir,

In a previous paper [1] we reported a gas chromatographic–electron-capture detection (GC–ECD) method for the quantitation of propafenone in biological fluids The major and active metabolite of propafenone, 5-hydroxypropafenone [2], is more acidic and polar than the parent drug Simultaneous measurement of both compounds is not possible without sacrificing the recovery of one or the other compound We have modified the previous GC–ECD assay to quantitate 5-hydroxypropafenone

EXPERIMENTAL

Materials and solutions

Propafenone, 5-hydroxypropafenone and Li-1548 (internal standard) were supplied by Knoll (Markham, Canada). Dichloromethane and 2-propanol (distilled in glass) were purchased from Caledon Labs (Georgetown, Canada) Other chemicals were the same as reported previously [1]

The 5-hydroxypropafenone hydrochloride and internal standard solutions were prepared by dissolving these compounds in methanol–water (1 9) (0 1 mg/ml, equivalent to base) and dilution to the desired concentrations

Instrumentation and analysis

The instrumentation for the analysis of 5-hydroxypropafenone was the same as previously reported for propafenone [1] Modifications of the GC conditions and the extraction and derivatization procedures are shown in Table I

0378-4347/89/$03 50

TABLE I

COMPARISON OF THE EXTRACTION, DERIVATIZATION AND ANALYSIS OF PROPAFENONE AND 5-HYDROXYPROPAFENONE

	Propafenone	5-Hydroxypropafenone
GC conditions		
Initial column temperature	220°C	205°C (for 0.8 min)
Temperature programming rate	4°C/min	3°C/min
Final column temperature	270°C	265°C
Extraction		
Solvent	Benzene	Toluene–dichloromethane–2-propanol (7:3:1)
Alkali solutions	1 *M* Sodium hydroxide (0.5 ml) 5 *M* Sodium hydroxide (0.5 ml)	0.1 *M* Sodium carbonate (0.15 ml) 5 *M* Potassium carbonate (0.5 ml)
Derivatization	Heptafluorobutyric anhydride (HFBA) (50 μl)	HFBA (20 μl)
Removal of excess HFBA	Evaporation	Mix with 0.5 ml of phosphate buffer (pH 6.0)

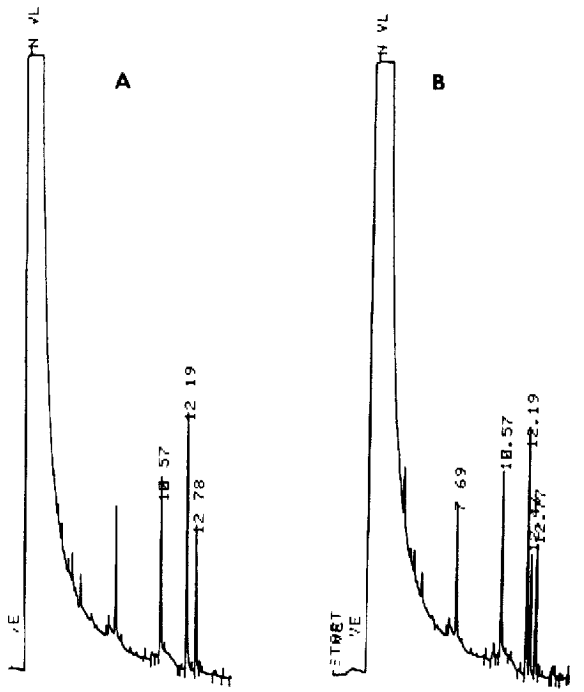

Fig 1 Chromatograms of (A) blank serum spiked with 5-hydroxypropafenone (0 02 μg, retention time 12 78 min) and Li-1548 (0 08 μg, retention time 12 19 min) and (B) chromatogram of serum sample from a volunteer receiving propafenone (retention time 12 47 min) spiked with Li-1548 (0 08 μg)

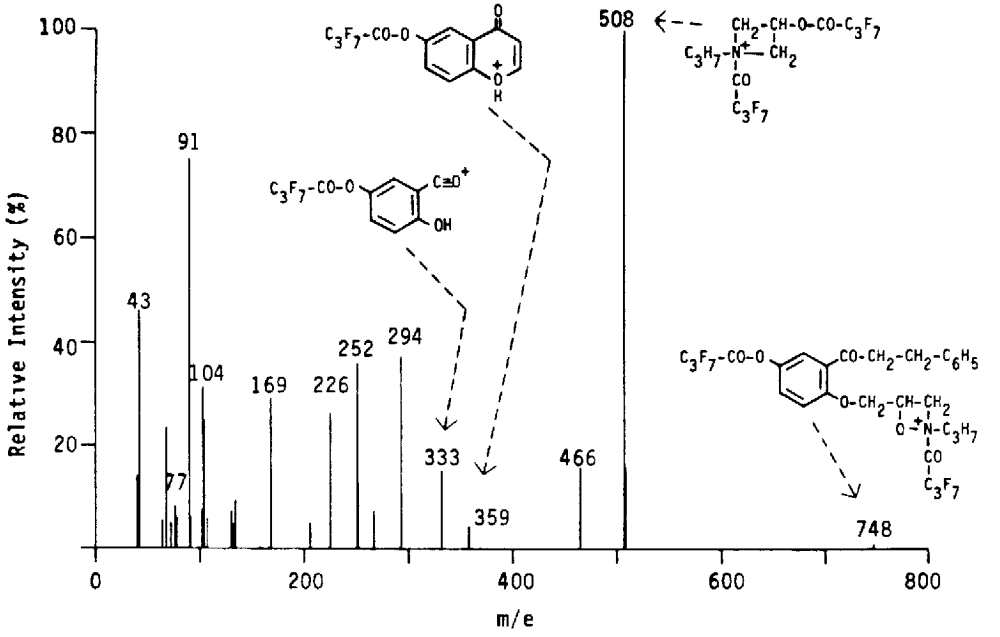

Fig 2 EI mass spectrum and selected proposed fragmentation structures of the HFB derivative of 5-hydroxypropafenone

RESULTS AND DISCUSSION

Fig. 1A and B show a chromatogram of blank serum spiked with 5-hydroxypropafenone and a chromatogram from serum obtained from a volunteer who received propafenone (2 h after a 300-mg oral dose), respectively. Although both propafenone and 5-hydroxypropafenone were measured (Fig 1B), the previous method [1] should be used for propafenone quantitation since the recovery of propafenone, using the modified extraction and derivatization procedure, is lower

This is the first GC–ECD method reported for the analysis of 5-hydroxypropafenone Linearity was observed over the range 2 5–50 ng/ml. A representative calibration curve was described by the regression equation $y = 0\ 023x + 0.052$ ($r = 0\ 996$). The coefficient of variation was less than 3% over the concentration range studied The recovery of 5-hydroxypropafenone was ca 88%

The heptafluorobutyryl (HFB) derivative of 5-hydroxypropafenone showed a fragmentation pattern similar to the HFB derivative of propafenone [1] The molecular ion (*m/e* 945) was not observed in the electron-impact mass spectrum Ions at *m/e* 333, 359 and 748 were characteristic of 5-hydroxypropafenone (Fig 2)

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