GLC Determination of Bromperidol in Tablets

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Received February 2, 1981, from the *Research Laboratories, Ortho Pharmaceutical Corporation, Raritan, NJ 08869.* Accepted for publication May 14, 1981. *Present address: Cilag-Ltd., Schaffhausen, Switzerland.

Abstract □ A GLC procedure for the determination of bromperidol in 3-, 6-, and 10-mg tablets is described. The method is stability indicating in the presence of compounds structurally related to bromperidol and stress reaction products of bromperidol.

Keyphrases □ Bromperidol—GLC determination in tablets □ GLC—method for determining bromperidol in tablets □ Tranquilizers—bromperidol, determination by GLC, tablets

Bromperidol is a new, potent, neuroleptic formulated in 3-, 6-, and 10-mg tablets; its chemical structure is similar to haloperidol. Analytical methods for haloperidol have included TLC (1), paper chromatography (2), spectrophotometry (3–5), titrimetry (6), polarography, and gas chromatography (7, 8), but only limited resolution data have been submitted for these methods. A TLC and a high-performance liquid chromatographic method for the determination of bromperidol were reported (9). These procedures are selective for bromperidol in the presence of three possible impurities and several other butyrophenones, but resolution from stress reaction products was not demonstrated, limiting data to support stability-indicating properties.

This paper describes a rapid, stability-indicating GLC procedure applicable to the determination of bromperidol in 3-, 6-, and 10-mg tablets. Selectivity in the presence of stress reaction products of bromperidol and other structurally related compounds is demonstrated.

EXPERIMENTAL

Reagents and Chemicals—Reagent grade octacosane (internal standard), cyclohexane, ethyl acetate, and sodium hydroxide were used.

Instrumentation and Apparatus—The gas chromatograph[1] was equipped with an autosampler, a flame-ionization detector, and on-column injection. A laboratory data system[2] was used to carry out peak integration or peak height measurement. However, peak heights could also be determined manually using an attenuation setting of 4×10^3 in combination with a 10-mv strip-chart recorder (1.5 min/cm).

Chromatographic Conditions—A 0.9-m (3 ft) × 2-mm i.d. glass column, packed with 3% SE-30 on 80–100-mesh Gas Chrom Q[3], was conditioned for 24 hr at 240° with a helium flow of ~30 ml/min. For sample determinations, the column temperature was adjusted to 230° with a detector temperature of 300° and an injection port temperature of 250°. The carrier gas was helium at a flow rate of 40 ml/min. Oxygen and hydrogen were maintained at 240 and 35 ml/min, respectively.

Preparation of Internal Standard—Approximately 100 mg of octacosane was accurately weighed into a 100-ml volumetric flask, dissolved in 10.0 ml of cyclohexane, and diluted to volume with ethyl acetate (for a concentration of 1 mg/ml).

Preparation of Bromperidol Standard Solution—Approximately 24 mg of a suitable bromperidol reference standard was accurately weighed into a stoppered 50-ml centrifuge tube and dissolved in 20.0 ml of the internal standard solution. Then 20.0 ml of 0.1 *M* NaOH was added. After shaking for 30 min, the mixture was centrifuged to ensure phase separation.

[1] Hewlett-Packard model 5710A.
[2] Hewlett-Packard model 3354.
[3] Supelco, Bellefonte, Pa.

Table I—Determination of Bromperidol in the Presence of Placebo Tablets (n = 6)

Amount Added, mg	Average Amount Found, mg	Recovery, %	*RSD*, %	Range, mg
3.010	3.037	100.9	0.2	3.028–3.043
6.021	6.155	102.2	0.2	6.112–6.208
10.11	10.21	101.0	1.2	10.00–10.33

Table II—Maximum Error at 90 and 110% of Label Claim Resulting from the Use of a Single Standard (100% Claim) Instead of a Calibration Curve

Formulation, mg	Maximum Error, % 90% of Label	110% of Label
3	0.21	−0.18
6	−0.02	0.07
10	0.69	−0.56

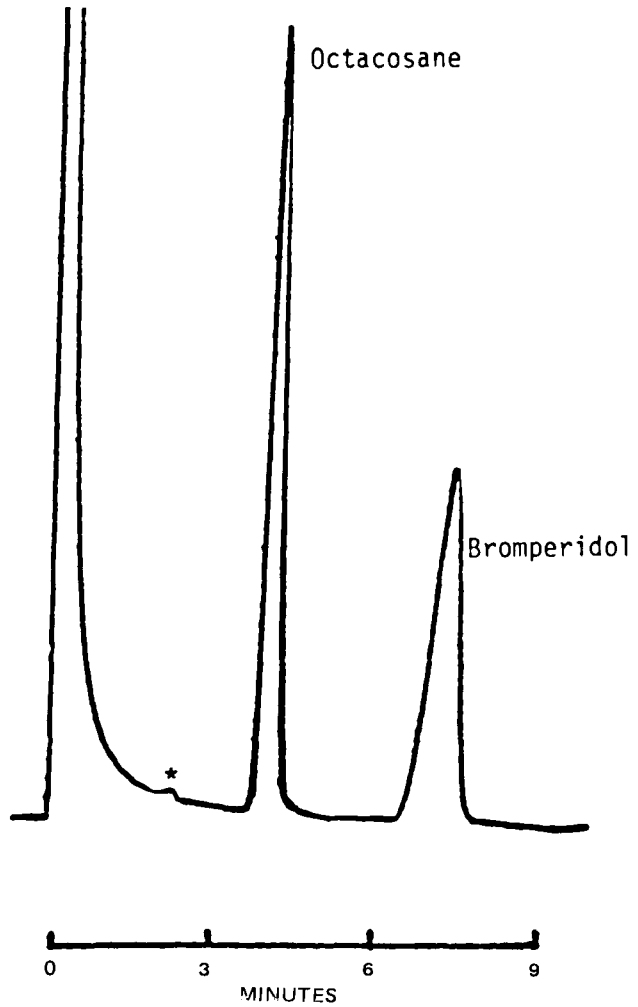


Figure 1—*Typical chromatogram of octacosane (1 mg/ml), and bromperidol (1.2 mg/ml). (*Impurity in octacosane.)*

0022-3549/82/0100-0112$01.00/0

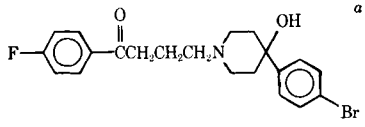
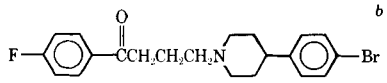
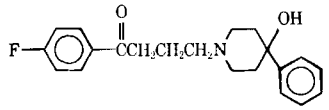
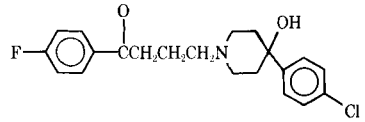
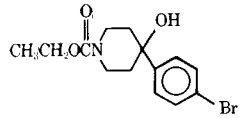
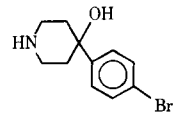
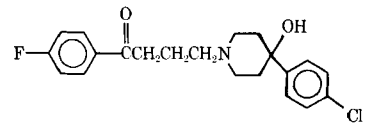
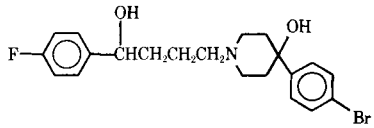
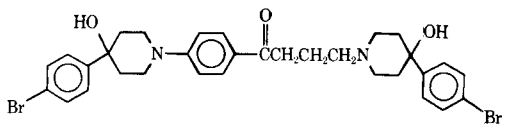
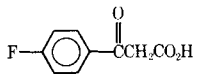
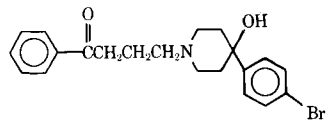
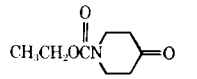
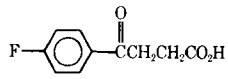
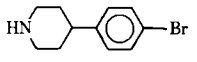
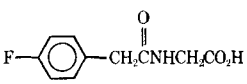
Table III—Determination of Bromperidol in Bromperidol Tablets by GLC and by Spectrophotometry

Experimental Lot	Whole Tablet GLC Determination Using Peak Areas: Average Bromperidol, mg/120-mg tablet	*n*	*RSD*, %	Range, mg	Ground Composite GLC Determination (20 Tablets) Using Peak Areas: Average Bromperidol, mg/120-mg tablet	*n*	*RSD*, %	Range, mg	UV Determination, Average Bromperidol, mg/120-mg Tablet
1	3.05	6	0.3	3.04–3.07	3.03	6	0.3	3.02–3.05	3.03
2	6.10	6	1.1	6.03–6.22	6.20	6	0.5	6.14–6.23	6.06
3	10.19	10	0.4	10.14–10.25	—	—	—	—	9.95

Table IV—Comparison of Determinations of Bromperidol in Tablets by GLC Based on Peak Heights to Similar Determinations Based on Peak Areas

3-mg Tablets (Lot 1)		6-mg Tablets (Lot 2)		10-mg Tablets (Lot 3)	
Peak Height	Peak Area	Peak Height	Peak Area	Peak Height	Peak Area
3.04	3.04	6.37	6.14	10.0	9.88
2.99	3.05	6.20	6.22	10.2	9.99
3.02	3.05	6.18	6.22	10.2	10.1
3.03	3.07	6.12	6.20	10.2	10.0
3.05	3.05	6.19	6.22	10.2	9.89
3.08	3.05	6.12	6.22	10.2	9.87
Average 3.04	3.05	6.20	6.20	10.2	9.96
RSD, % 1.0	0.3	1.5	0.5	0.8	0.9

Table V—Relative Retention Time and Weight Response of Compounds Structurally Related to Bromperidol

Compound	Relative Retention Time	Relative Weight Response
 [a]	1.00	1.00
 [b]	0.57	1.47
	0.80	0.95
	0.41	1.11
	1.22	0.34
	0.15	0.90
	0.05	0.53
	0.74	0.93
	0.90	0.45
	NO [c]	—
	NO	—
	NO	—
	NO	—
	NO	—
	NO	—
	NO	—

[a] Bromperidol. [b] Octacosane. [c] Not observed.

RESULTS AND DISCUSSION

Accuracy and precision were demonstrated in two ways. First, placebo tablets in the presence of known concentrations of bromperidol were assayed by the desired procedure. Results obtained for six replicate determinations of synthetic preparations representative of 3-, 6-, and 10-mg bromperidol tablets show average recoveries of 100.9 ± 0.2, 102.2 ± 0.2, and 101.0 ± 1.2%, respectively (Table I). No interference was encountered from the excipients. A typical chromatogram is shown in Fig. 1.

For each formulation, the area response was linear with concentration. Table II presents the maximum error at 90 and 110% of label claim resulting from the use of a single standard representative of 100% of label claim instead of a calibration curve. Since the maximum error was within the precision of the method, the use of a standard at the nominal concentration in place of a series of concentrations was justified.

Accuracy and precision were also evaluated by comparing results obtained using the described procedure with results obtained by an independent UV spectrophotometric procedure. In the latter method, the

drug was extracted from the tablet matrix using isopropyl alcohol–0.1 *M* hydrochloric acid (9:1) and diluted to a final bromperidol concentration of 15 μg/ml with isopropyl alcohol–0.1 *M* hydrochloric acid. The bromperidol present in the sample was calculated by comparing the absorbance of the sample at 242 nm to a similarly prepared solution of standard at a known concentration. Table III shows the agreement between results obtained by both procedures using actual 3- and 6-mg bromperidol tablets. Equivalent results were obtained independent of whether ground composites or whole tablets were assayed. Table III also shows data collected on representative 10-mg tablet lots.

The data in Table IV demonstrate that peak heights can be used interchangeably with peak areas for quantitation. These data were determined using individual determinations of 3-, 6-, and 10-mg tablets calculated by both techniques. Overall results as well as differences between individual results were within experimental error.

To demonstrate the stability-indicating properties of the method, the chromatographic response of several compounds structurally related to bromperidol were compared to bromperidol and octacosane (the internal standard) to establish resolution properties. Each compound was dissolved in ethyl acetate–cyclohexane (9:1), and the resulting solutions were chromatographed. Besides bromperidol, only seven compounds produced discernible peaks. All compounds were chromatographed individually; Table V presents the relative retention times and weight responses relative to bromperidol (the first compound). Compounds not observed were concluded to have either eluted with the solvent front or been strongly retained by the packing. However, none of the compounds interfered with the determination of bromperidol.

To defend further the resolution properties of the method and, therefore, the stability-indicating properties, a weighed amount of bromperidol drug substance was heated in a capillary tube at 260° (melting point of bromperidol is 158°). After ~30 min at this extreme temperature, the sample was partially discolored near the top of the tube; analysis for bromperidol indicated a drug loss of ~25%. However, no extra peaks were observed in the resulting chromatogram. Examination of the analytical regions of the resultant chromatogram using GLC–mass spectrometry indicated that thermal stress reaction products of bromperidol did not elute with the drug or internal standard.

In addition to the thermal stress experiment, an accurately weighed 25-mg sample of bromperidol was mixed with 2 ml of 30% hydrogen peroxide and allowed to stand for 1 hr. The sample was mixed with 10 ml of methanol to consume excess peroxide and evaporated to dryness under nitrogen. GLC analysis of the residue indicated as little as 10% intact drug remaining. Examination of the oxidized sample of bromperidol by GLC–mass spectrometry in the analytical regions indicated that reaction products of bromperidol did not elute with the internal standard or bromperidol.

This GLC method is specific for bromperidol in the presence of structurally related compounds and stress reaction products of bromperidol. In addition, the procedure is accurate, reproducible, and suitable for stability or release analyses of bromperidol tablets.

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ACKNOWLEDGMENTS

The work of Mr. C. Shaw, who performed the GLC–mass spectrometry, is gratefully acknowledged.

High-Performance Liquid Chromatograhic Determination of Metoprolol in Plasma

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Received November 28, 1980, from the **Heymans Institute of Pharmacology and the ‡Department of Cardiology, University of Gent Medical School, De Pintelaan 135, B-9000 Gent, Belgium.* Accepted for publication May 20,1981.

Abstract □ A high-performance liquid chromatographic method is presented for the determination of metoprolol in human plasma. Metoprolol was extracted from plasma by a single extraction procedure with 4-methylpropranolol as the internal standard. Chromatography was done on a reversed-phase column with fluorescence detection. The minimum detectable concentration was 5.0 ng/ml of plasma. The standard curve was linear in the range evaluated, 10–300 ng/ml. The within-run coefficient of variation was 2.3–6.0%, and the day-to-day variation was 6.8%. The method is free from interference by major metoprolol metabolites.

Keyphrases □ Metoprolol—high-performance liquid chromatographic analysis in plasma □ β-Adrenergic blocking agents—metoprolol, high-performance liquid chromatographic analysis in plasma □ High-performance liquid chromatography—analysis of metoprolol in plasma

Metoprolol is a cardioselective β-adrenergic blocking agent used in the treatment of hypertension and angina. Analysis of metoprolol in plasma has been performed with electron-capture GLC after derivatization (1, 2), and a GC–mass spectroscopy method was recently developed (3).

This report describes a high-performance liquid chromatographic (HPLC) method for the determination of metoprolol in plasma using reversed-phase ion-pair chromatography with fluorescence detection. The same approach has been used for other β-adrenergic blocking drugs such as propranolol (4, 5), atenolol (6, 7), and sotalol (8).

EXPERIMENTAL

Reagents—The following were used: metoprolol tartrate[1], α-hydroxymetoprolol[1], and *O*-desmethylmetoprolol[1] as the *p*-hydroxybenzoic acid salts; and 4-(2-hydroxy-3-isopropylaminopropoxy)phenylacetic acid[1], 2-hydroxy-3-(4-methyoxyethylphenoxy)propanoic acid[1], and 4-methylpropranolol[2] as the hydrochloride salts. Methanol[3] was HPLC grade. 1-Heptanesulfonic acid[4] in acetic acid was used. Glass double-distilled water was passed through a 0.45-μm filter[5]. Methylene chloride was reagent grade and distilled just before use.

Instrument Conditions—The microprocessor-controlled high-per-

[1] Hässle, Mölndal, Sweden
[2] Imperial Chemical Industries, Macclesfield, England.
[3] Burdick & Jackson Laboratories, Muskegon, Mich.
[4] Reagent B-7, Waters Associates, Milford, Mass.
[5] Type HA Millipore Corp., Bedford, Mass.

0022-3549/82/0100-0114$01.00/0