GLC Assay of Meprobamate and Related Carbamates

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Abstract □ A specific and rapid GLC method was developed for the determination of meprobamate in bulk powder form and in tablets. The method utilizes a simple extraction procedure followed by chromatography on a 3.8% OV-17 column, with the injection port and column oven at the same temperature. The use of tybamate as the internal standard permits the analysis of the related propanediol dicarbamates, carisoprodol and mebutamate, with no modifications necessary.

Keyphrases □ Meprobamate—GLC analysis in powder and tablets □ Tybamate—internal standard for GLC analysis of meprobamate, carisoprodol, and mebutamate □ GLC—analysis, meprobamate, carisoprodol, and mebutamate

Meprobamate (2-methyl-2-propyl-1,3-propanediol dicarbamate) is the most well-known member of a family of propanediol dicarbamates possessing tranquilizing and skeletal muscle relaxant properties. Some others are tybamate [2-(hydroxymethyl)-2-methylpentyl butylcarbamate carbamate], carisoprodol (*N*-isopropyl-2-methyl-2-propyl-1,3-propanediol dicarbamate), and mebutamate (2-*sec*-butyl-2-methyl-1,3-propanediol dicarbamate). Methods for the quantitative analysis of these compounds are based on the chemistry of the carbamate moiety. They include aqueous acid hydrolysis followed by volumetric determination of the liberated ammonia (1) and nonaqueous alkaline solvolysis with the formation of inorganic cyanate (2, 3). Methods based on the acid-catalyzed condensation with an aldehyde (4, 5) and the ability of hypochlorite to *N*-chlorinate unsubstituted carbamate nitrogen (6) have been reported. Spectral methods based on IR, near IR, and NMR absorption also have been reported (7–9).

Several GC methods have been proposed. Those reported are specific for a particular carbamate and are generally adequate for biochemical analysis such as the determination of blood levels (10). However, they are less satisfactory for the quantitative, routine analysis of tablet dosage forms and bulk powders because of decomposition in the GC system (11–14). Poor resolution or efficiency (15) or the need for derivative formation for successful quantitative analysis also has been cited (16).

This paper describes the rapid quantitative GLC determination of meprobamate under conditions giving no evidence of degradation. A low boiling solvent, with the column oven and injection port at the same temperature, on-column injection into an all-glass system, and the proper internal standard are all essential to the determination. Data demonstrating linearity of response with changes in concentration, reproducibility, and completeness of recovery are presented.

Table I—Meprobamate: Tablet and Bulk Powder Analysis

Run Number	Tablets, mg. Found[a]	Bulk Powder, % Found
1	388.9	99.45
2	386.0	99.91
3	388.2	99.26
4	386.0	98.91
5	388.7	99.85
Mean	387.6 (389.6)[b]	99.48
σ	±1.4	±0.42

[a] Label claim, 400 mg./tablet. [b] Analysis of meprobamate by independent method (17). Grand average of 20 determinations on individual tablets.

EXPERIMENTAL

Operational Parameters—A gas chromatograph[1] with hydrogen flame-ionization detectors was used. The column was glass, with a U-type configuration, 76 cm. long by 3 mm. i.d., packed with 3.8% methyl phenyl silicone fluid on a silanized diatomaceous earth support, 80–100 mesh. The column was conditioned by heating at 250°, for 1 hr. with carrier gas flow. After cooling to room temperature, the carrier gas flow was stopped and the column was heated at 330° for 4 hr. After cooling again to room temperature, the column was conditioned for 18 hr. at 250° with carrier gas flow. The operating parameters were: column temperature, 170°; detector temperature, 260°; injection port temperature, 170°; and carrier gas, nitrogen at 45 ml./min. Hydrogen and air flows were adjusted to obtain maximum efficiency as recommended by the manufacturer; the range selector was set at 10 and attenuation was adjusted to obtain at least a 50% recorder scale response for all peaks[2].

Reagents and Chemicals—Liquid phase methyl phenyl silicone fluid (OV-17) and silanized diatomaceous earth support (Gas Chrom Q) were used as supplied[3]. Meprobamate reference standard and tybamate reference standard were the USP and NF standards, respectively. All solvents were of GC grade.

Standard Preparation—*Meprobamate Standard Solution*—USP meprobamate reference standard was accurately weighed and diluted with methylene chloride to a concentration of 1.0 mg./ml.

Tybamate Internal Standard Solution—NF tybamate reference standard was prepared to contain 2.0 mg./ml. in methylene chloride.

Working Standard Solution—A 2.0-ml. aliquot of the meprobamate standard solution was evaporated to dryness in a suitable glass vial and reconstituted with a 1.0-ml. aliquot of the tybamate internal standard solution. A 0.8–1.2-μl. sample was injected into the chromatograph and allowed to elute for about 12 min. Under the stated operational conditions, the relative retention time of

[1] Hewlett-Packard model 402.
[2] All peak areas were measured with a Disc integrator.
[3] Applied Science Laboratories, State College, Pa.

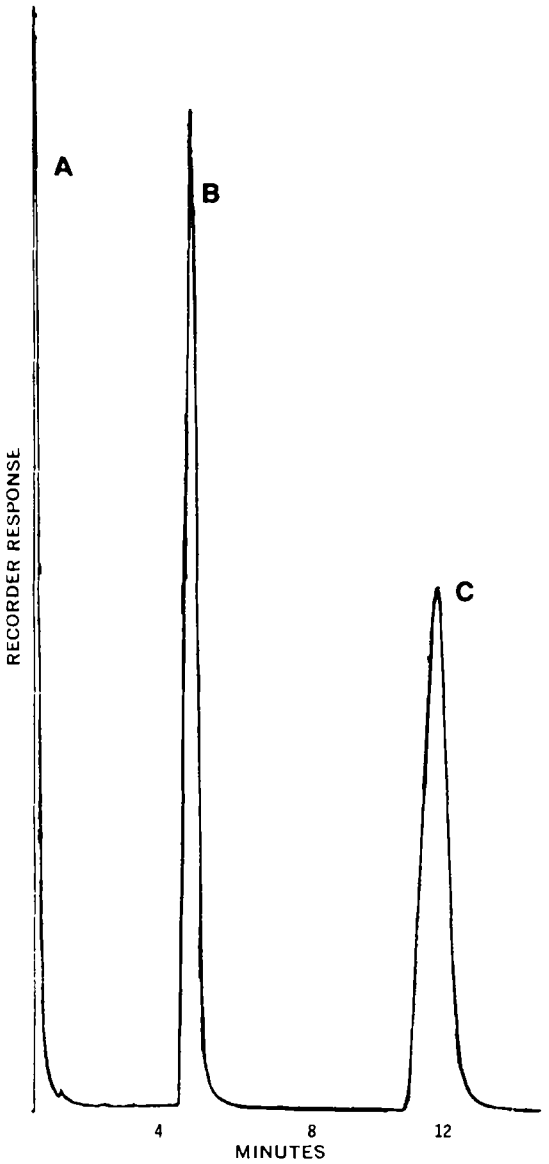

Figure 1—*Standard chromatogram showing relative retention times of meprobamate and the internal standard tybamate. Key: A, solvent; B, meprobamate; and C, tybamate.*

meprobamate was 0.38 relative to the internal standard. A typical standard chromatogram is shown in Fig. 1.

Sample Preparation—The preparation of a bulk powder sample for analysis was carried out in a manner identical to the preparation of the meprobamate and working standard solutions. Tablet dosage forms were ground to a fine, uniform powder; an accurately weighed sample, containing about 40 mg. of meprobamate, was transferred to a 50-ml. glass vial and methanol (20.0 ml.) was added. The vial was capped securely and shaken vigorously for 30 min. After centrifugation at moderate speed, a 1.0-ml. aliquot of the clear supernatant fluid was transferred to a suitable vial and evaporated to dryness, and the residue was dissolved in 1.0 ml. of the tybamate internal standard solution. A 0.8–1.2-μl. injection was made and allowed to elute for about 12 min.

RESULTS AND DISCUSSION

Tybamate was selected as the internal standard because of its excellent separation from trace contaminants of meprobamate, its functional group similarity to meprobamate and related dicarbamates, and its stability under the analytical conditions. In addition to its use for the analysis of meprobamate, it may be used as the internal standard for the analysis of carisoprodol (relative retention time to tybamate is 0.44) or mebutamate (relative retention time to tybamate is 0.60) using the operating conditions and parameters already described.

Table II—Meprobamate Recovery Study

Amount Present, mg.	Experimentally Found, mg.	Recovery, %
40.6	41.2	101.5
39.6	39.4	99.5
40.4	40.5	100.2
40.7	41.3	101.5
41.4	41.8	101.0
Mean		100.7

Standard solutions of meprobamate, ranging from 25 to 125% of the concentration levels to be expected in a typical analysis, were chromatographed; a plot of the ratio of areas *versus* concentration (Fig. 2) shows that the chromatographic response is linear over the entire range.

Replicate determinations of a commercial tablet containing a labeled amount of 400 mg. meprobamate and a production batch of bulk powder (Table I) were made. The standard deviations calculated indicate excellent precision for the analytical procedure.

A recovery study of the procedure was made by directly adding known quantities of meprobamate to a standard placebo tablet formulation and dry mixing before extraction. The results are shown in Table II. The standard chromatogram (Fig. 1) indicates that there is little, if any, degradation of meprobamate or tybamate as it passes through the chromatographic system. This is due, in

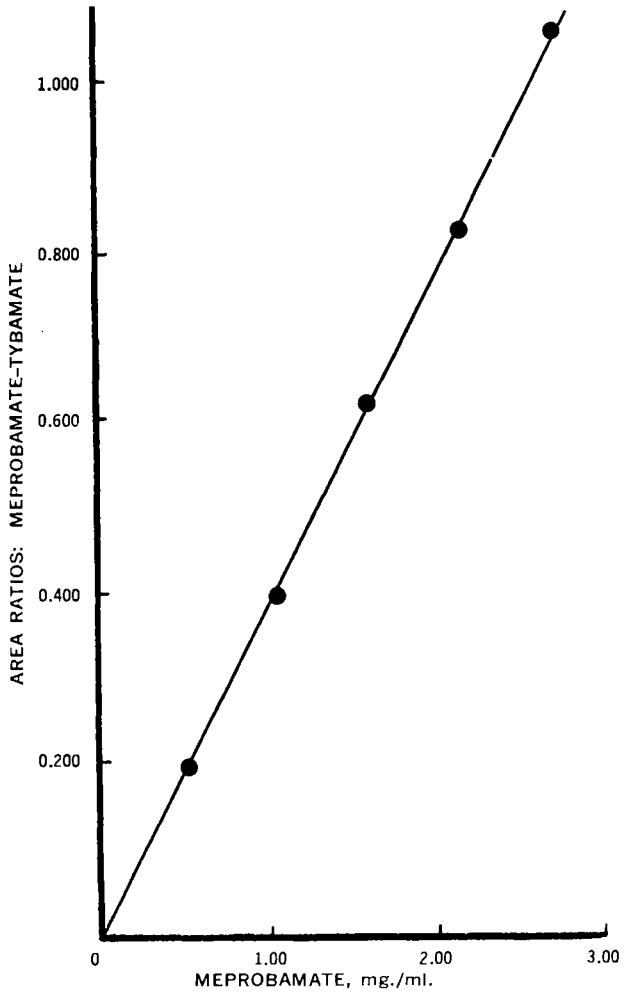

Figure 2—*Linearity of response (ratio of areas) with concentration. The column is 3.8% OV-17 on Gas Chrom Q, length 76 cm., temperature 170°. Concentration is in milligrams per milliliter, with the internal standard as a constant.*

part, to the temperature of the injection port (ambient, relative to the oven temperature). As the injection port temperature was increased 20° or more over the oven temperature, the appearance of a peak on the trailing edge of the solvent front became pronounced. This peak was identified as 2-methyl-2-propyl-3-hydroxypropyl carbamate, a trace impurity in meprobamate and occasionally detected by TLC or GC methods. Holch and Gjaldbaek (11) confirmed that it also arises as a detectable degradation compound when the injection port is greater than 210°. Another commonly encountered compound is 2-methyl-2-propyl-1,3-propanediol. The "diol" is usually lost in the solvent peak and does not interfere in the analysis. Other known trace contaminants of meprobamate were tested and showed retention times that were either greater or lesser than meprobamate, thereby assuring no interference in the analytical procedure for the determination of meprobamate.

SUMMARY

A specific and rapid GLC method has been developed for the determination of meprobamate in bulk powder form and in tablets. The method for tablets utilizes a simple extraction procedure and subsequent combination with an internal standard. The use of tybamate as the internal standard permits the analysis of the related propanediol dicarbamates, carisoprodol and mebutamate, with no modifications necessary. Degradation of meprobamate in the GLC system is not apparent. Conditions are outlined for the operational parameters and extraction procedure.

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Determination of Isomeric Composition of Amphetamine Mixtures from Melting Points of Monohydrogen Succinate Salts

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Abstract □ A rapid and simple method for the identification of particular amphetamines and the determination of their isomeric composition is presented. The method involves the preparation of the monohydrogen succinate salt and uses the melting point of this derivative in the accurate determination of the isomeric composition of the amphetamine. Application of this procedure to actual samples as shown has affirmed the utility and validity of this method.

Keyphrases □ Amphetamine sulfate isomer mixtures—determination of isomeric composition from melting point of monohydrogen succinate salt □ Dextroamphetamine sulfate—determination in amphetamine sulfate mixtures from melting point of monohydrogen succinate salt □ Isomer mixtures, amphetamine sulfate—determination of components □ Succinic acid—amphetamine sulfate salt formation, determination of isomeric composition

The difference in the physiological response to *d*-amphetamine (dextroamphetamine) as compared to the *l*-stereoisomer has indicated the need for an isomer-measuring analytical procedure. Welsh (1, 2) discussed the problem from a phase diagram point of view and introduced a procedure, accurate to about 1%, involving the formation of the acetyl derivative and the determination of melting points. A recent paper (3) reviews the literature and describes a procedure for determining the optical isomer composition from the melting point of the benzoyl derivative.

The method reported here involves the preparation of the monohydrogen succinate salt of amphetamine and uses the melting point of this derivative to deter-