Quantitative Determinations of Phenol and Resorcinol in Pharmaceutical Dosage Forms by High-Pressure Liquid Chromatography

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Abstract □ The quantitative determinations of phenol in phenolated calamine lotion USP and of phenol and resorcinol in phenol–resorcinol-boric acid solution by high-pressure liquid chromatography are reported. The procedures are simple, rapid (no special preliminary treatment is required), and accurate. There is no interference from other ingredients of the lotion (bentonite magma, calamine, and zinc oxide) or solution (acetone and boric acid).

Keyphrases □ Phenol—high-pressure liquid chromatographic analysis, pharmaceutical dosage forms □ Resorcinol—high-pressure liquid chromatographic analysis, pharmaceutical dosage forms □ High-pressure liquid chromatography—analysis, phenol and resorcinol, pharmaceutical dosage forms □ Preservatives—phenol, high-pressure liquid chromatographic analysis, pharmaceutical dosage forms □ Keratolytic agents—resorcinol, high-pressure liquid chromatographic analysis, pharmaceutical dosage forms

No method is available for the direct quantitative determination of phenol and resorcinol in each other's presence. A reported method (1) required a combined assay on both compounds by a bromometric technique and then division of the data based on the molar ratio obtained from the NMR spectroscopic analysis.

The purpose of this investigation was to develop a simple, rapid, and accurate method for the direct quantitative determination of both phenol and resorcinol in a pharmaceutical dosage form. The developed high-pressure liquid chromatographic (HPLC) method was successfully applied to the quantitative determination of phenol in phenolated calamine lotion USP.

EXPERIMENTAL

Chemicals and Reagents—All chemicals and reagents were ACS, USP, or NF quality and were used without further purification.

Apparatus—A high-pressure liquid chromatograph[1] equipped with a UV detector (254 nm) and a recorder[2] was used.

Column—A column (30 cm × 4 mm i.d.) of a very nonpolar packing material[3] consisting of a monomolecular layer of octadecyltrichlorosilane permanently bonded by silicon–carbon bonds was used.

Chromatographic Conditions—The chromatographic solvent was 0.02 M NH_4HCO_3 in 10% by volume of methanol in water[4] (pH 7.67). The temperature was ambient, and the solvent flow rate was 3.0 ml/min (at an inlet pressure of approximately 2200 psig). The detector was set at a sensitivity of 0.16 absorbance unit full scale, and the chart speed was 30.5 cm (12 in.)/hr.

Preparation of Solutions—An aqueous standard solution of phenol (45.0 mg/100 ml), resorcinol (100.0 mg/100 ml) or a combination of phenol and resorcinol was prepared using the simple solution method. Phenol-resorcinol-boric acid solution was prepared according to a procedure reported earlier (1). The phenolated calamine lotion was prepared according to directions in the USP (2).

Assay Procedure for Phenol and Resorcinol in Phenol-Resorcinol-Boric Acid Solutions—Dilute 10.0 ml of the solution to 1000.0 ml with water, mix, and inject 10–20 µl.

[1] Waters ALC 202 equipped with U6K universal chromatograph injector.
[2] Omniscribe 5213-12 equipped with an integrator.
[3] Waters µBondapack-C18, Catalog No. 27324.
[4] Must be used within 3 days since pH changes on standing.

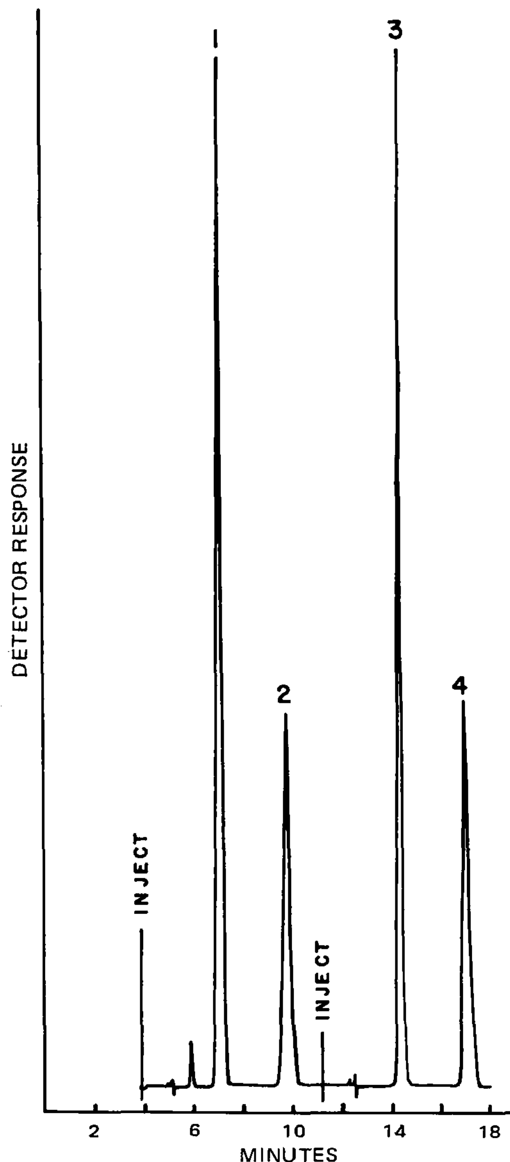

Figure 1—*HPLC analysis of phenol and resorcinol in combination (for chromatographic conditions, see text). Peaks 1 and 2 are resorcinol and phenol, respectively, from an assay sample; peaks 3 and 4 are resorcinol and phenol, respectively, from a standard sample.*

Assay Procedure for Phenol in Phenolated Calamine Lotion USP—Shake the lotion thoroughly and dilute 5.0 ml to 100 ml with water. Mix, filter, reject the first 8–10 ml of the filtrate, collect the clear filtrate, and inject 10–20 µl.

For the purpose of comparison, inject an identical volume of the appropriate standard solution after the assay solution is eluted.

Calculations—Since preliminary investigations indicated that the peak areas of phenol and resorcinol were directly related to the concentrations, the results were calculated using:

$$\frac{A_a}{A_s} \times 100 = \text{percent of label claim} \qquad \text{(Eq. 1)}$$

Table I—Assay Results on Phenol and Resorcinol

Sample	Percent Results[a] on Phenol	Resorcinol
1[b]	99.6	100.2
2[b]	98.7	99.4
3[c]	100.5	0.0
Average deviation	±2.4	±1.8

[a] Average of three. [b] Phenol–resorcinol–boric acid solution. [c] Phenolated calamine lotion.

where A_a = peak area of the assay sample, and A_s = peak of the standard sample.

The results are presented in Table I, and the chromatogram is presented in Fig. 1.

DISCUSSION

The assay results (Table I) indicate that phenol and resorcinol in combination can be separated (Fig. 1) and assayed accurately. The procedure developed for the quantitative determinations of phenol and resorcinol by HPLC is very simple and rapid (no special preliminary treatment is required). No interference from other ingredients of the phenol–resorcinol–boric acid solution (acetone and boric acid) or phenolated calamine lotion USP (bentonite magma, calamine, and zinc oxide) was noted. The method provides direct results on phenol and resorcinol rather than determining one or the other by difference.

REFERENCES

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Electron-Capture GC Determination of Subnanogram Amounts of Emepronium Bromide in Serum

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Abstract □ Barium peroxide in an acidic medium was utilized to increase the sensitivity in the benzophenone method for the determination of the quaternary ammonium compound emepronium bromide. The method is comprised of ion-pair extraction, oxidation, and quantitative determination of benzophenone by electron-capture GC. By employing small extraction and reaction volumes, the method was used in the 0.2–8-ng/ml range with a relative standard deviation of 2.5% at the 1-ng/ml level. The application of the method to human serum samples after a single oral dose demonstrated that the elimination phase for emepronium in serum had a half-life of 7–11 hr.

Keyphrases □ Emepronium bromide—electron-capture GC analysis in human serum, elimination profile described □ GC, electron capture—analysis, emepronium bromide in human serum □ Elimination profile—emepronium bromide, electron-capture GC analysis in human serum □ Anticholinergic agents—emepronium bromide, electron-capture GC analysis in human serum, elimination profile described

Emepronium bromide[1] [(3,3-diphenyl-1-methylpropyl)dimethylethylammonium bromide], a quaternary ammonium compound with anticholinergic properties, was determined by electron-capture GC in biological samples after oxidation with chromic acid to benzophenone (1). The oxidation conditions as well as the extraction of emepronium were studied in detail. Special attention was paid to the interference from metabolites. The lower limit of detection of that method was about 1 ng/ml, and down to 3 ng/ml could be determined with acceptable precision. The fate of emepronium bromide in dogs (2) and humans (3) was studied with this sensitive method.

From a pharmacokinetic point of view, there was a need for a method with a higher sensitivity to follow the fate of emepronium over a longer time. Subnanogram analysis of emepronium bromide could be realized when a study of barium peroxide oxidation in sulfuric acid revealed very low reagent blanks (4). The sensitivity was also increased by the use of small extraction and reaction volumes and by the employment of a purer internal standard (5). This paper describes the development of a barium peroxide oxidation procedure for emepronium and its application to the analysis of human serum samples for the drug after single oral doses.

EXPERIMENTAL

Apparatus and Glass Equipment—*GC*—A gas chromatograph[2] equipped with a tritium electron-capture detector operating in the dc mode was used. The glass column (1.5 m × 1.8 mm) was filled with 3% DC 560 and 0.3% polyethylene glycol[3]–terephthalic acid on 100–120-mesh Gas Chrom P, acid washed and silanized. The column temperature was 139°; the flow rate of the carrier gas, nitrogen, was 30 ml/min. The detector temperature was 155°, corresponding to 138° at the detector foil.

Heating Bath—The reaction was performed in a dry bath[4] at 114°. The reaction tubes were emersed in sand so that only the water phase was covered.

Glass Equipment—The extraction was performed in 15-ml centrifuge tubes with a tapered base of 0.3 ml. The oxidation was performed in glass tubes with a height of 10 cm and an inner diameter of 0.6 cm. The tubes had a restriction at the half-height, and no condenser was necessary with this arrangement.

Reagents and Chemicals—Barium peroxide[5], anhydrous powder,

[1] Cetiprin.

[2] Varian 1400.
[3] Carbowax, Union Carbide.
[4] Thermolyne.
[5] Matheson, Coleman and Bell.