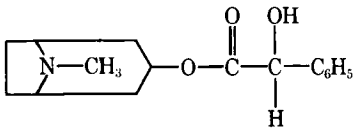
Stability Determination of Homatropine Hydrobromide by Direct Cerimetric Titration

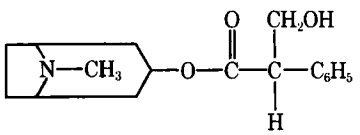
By LESTER CHAFETZ and ROBERT E. DALY

Using ceric nitrate as the titrant, the difference in titer between saponified and unhydrolyzed aliquots of homatropine solutions provides a measure of intact drug in the presence of its mandelic acid hydrolysis product. The titration is conducted on chilled solutions to circumvent interference by tropine, the other hydrolysis product, which is slowly oxidized by the titrant at room temperature. The proposed procedure is highly selective for the drug in the presence of its hydrolysis products, other tropine esters, and most other organic nitrogenous bases. The cerimetric titration method is rapid, absolute, and stability indicating.

HOMATROPINE IS A synthetic homolog of the solanaceous alkaloid atropine in which the tropic acid moiety of the ester alkaloid is replaced by *d,l*-mandelic acid. It is used in medicine as an ophthalmic anticholinergic agent, having the



Homatropine



Atropine

advantage over atropine of a shorter duration of activity (1). Since the hydrolysis products of homatropine, *i.e.*, tropine and mandelic acid, are inactive, the findings of Patel and Lemberger (2) that homatropine hydrolyzes at the pH of tears at several times the rate of atropine provide a rational basis for its shorter activity.

Patel and Lemberger (2) followed the rates of hydrolysis of homatropine under various experimental conditions by means of UV spectrophotometry after separation of intact mandelate ester from free mandelic acid by solvent extraction. Their scheme provides a valid stability method for homatropine, for the mandelic acid moiety comprises the UV chromophore. However, the procedure is somewhat tedious, and current practice requires the preparation or procurement of a reference standard for spectrophotometric comparison. Because the UV spectrum is benzenoid, the method is relatively nonselective.

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The assay for homatropine hydrobromide ophthalmic solution in USP XVII (3) does not discriminate between intact and hydrolyzed homatropine. The prescribed method is acetous perchloric acid titration of base extracted with chloroform from ammoniacal solution. (Measurements in the authors' laboratories—which will be described more fully in a future communication—indicate that tropine is well extracted from alkaline solution by chloroform. The partition coefficient, c_{CHCl_3}/c_{H_2O}, is 0.55.)

Chafetz and Gaglia (4) reported the direct titration of mandelic acid with ceric nitrate in dilute nitric acid and suggested application of the method to the assay of homatropine and other arylglycolate esters. The reaction was found to be highly selective for the free arylglycolic acids; their esters are not oxidized by ceric nitrate. Taken with quantitative data showing that homatropine is stable to hydrolysis in dilute acid and very labile at high pH (2), these observations afford a rational basis for an absolute assay of homatropine hydrobromide by direct cerimetric titration. A procedure for the determination of homatropine in the presence of its hydrolysis products and common preservatives and stabilizing agents is presented here and proposed for compendial use.

EXPERIMENTAL

Reagents and Supplies—Homatropine hydrobromide USP (Merck and Co., Inc.), chlorobutanol, and benzalkonium chloride (both USP) were used as received. The titrant was 0.05 *N* ceric nitrate, a 2.45% solution of ceric ammonium nitrate (G. Frederick Smith Co.) in 1 *N* nitric acid. It was standardized against primary standard ferrous ethylenediammonium sulfate (G. Frederick Smith Co.) as previously described (4). Nitroferroin T.S. was used as the end point indicator; it was prepared by dissolving 150 mg. of 5-nitro-1,10-phenanthroline in 15 ml. of freshly made aqueous 1.4% ferrous sulfate.

Assay—Dissolve an accurately weighed sample of homatropine hydrobromide in distilled water or

suitably dilute an accurately measured volume of homatropine hydrobromide ophthalmic solution to obtain a concentration of about 8 mg./ml. Transfer a 10.0-ml. aliquot of the solution or dilution to a 100-ml. beaker, add 5 ml. of 1 *N* sodium hydroxide, and heat the solution just to boiling on a hot plate. Add 10 ml. of 1 *N* nitric acid, dilute the solution to 50 ml. with water, and cool it to 0–5° in an ice bath. Concomitantly add 5 ml. of 1 *N* nitric acid to a second 10.0-ml. aliquot of the solution or dilution, dilute it to 50 ml. with water, and chill it in an ice bath. Add 1 drop of nitroferroin indicator to each of the beakers, and titrate the solutions to the disappearance of the pink color while stirring magnetically, using 0.05 *N* ceric nitrate as titrant from a 10-ml. buret. Similarly, titrate a blank consisting of 50 ml. of 0.1 *N* nitric acid and 1 drop of indicator, and make any indicated corrections. (The indicator requires acidity for a sharp color change.) Each milliliter of 0.05 *N* ceric nitrate is equivalent to 8.907 mg. of $C_{16}H_{21}NO_3 \cdot HBr$.

The titer of the saponified aliquot is a measure of the total amount of intact and hydrolyzed drug; the titer of the unsaponified aliquot affords a value for hydrolyzed ester. Thus, the difference in titers is equivalent to unhydrolyzed homatropine.

Stability Evaluations—No samples of homatropine hydrobromide ophthalmic solution were available to the authors for this study. In order to demonstrate the applicability of the method to partially degraded samples, solutions of the drug buffered at pH 9.95 were heated at 30° for periods of time monitored by means of a stopwatch, the saponification reaction was quenched by addition of nitric acid, and the samples were assayed for intact homatropine as described above. The apparent first-order rate constant obtained was compared with that calculated by the equation developed by Patel and Lemberger (2).

RESULTS AND DISCUSSION

Accuracy and Precision—A comparison of the results obtained by the proposed method with the USP XVII acetous perchloric acid titration method for homatropine hydrobromide is presented in Table I. An average of 99.3 ± 0.3% (*RSD*) was obtained by the cerimetric method. The precision was comparable to that previously reported (4) for mandelic acid in methenamine mandelate, and it is comparable to the result of 99.2 ± 0.33% (*RSD*) obtained in the USP assay. It may be noted that the cerimetric and USP methods measure different parts of the homatropine molecule. The titer for the unsaponified aliquot in the cerimetric determinations of homatropine hydrobromide bulk drug was the same as for the indicator blank, indicating no hydrolysis.

Selectivity—It has been noted previously (4) that, although ceric nitrate is a highly selective oxidant under the specified conditions, it is by no means specific. Tropine was found to be slowly oxidized at room temperature. Good values were obtained for the mandelic acid content of homatropine at room temperature, but the indicator end point was transient. This interference was circumvented by performing the titration on a chilled solution.

Chlorobutanol and benzalkonium chloride, two of the commonly used preservatives for ophthalmic solutions, were found to be unaffected by the ceric titrant. Many of the easily oxidized preservatives such as parabens could be removed by solvent extraction from the acidified solutions if they were present.

Table I—Comparison of USP and Cerimetric Methods for Homatropine

	% $C_{16}H_{21}NO_3 \cdot HBr$
USP XVII method	99.6, 99.1, 99.5, 98.7, 99.3, 99.0
Proposed cerimetric titration	98.8, 98.8, 99.6, 99.0, 99.3, 99.6
	99.6, 99.3, 99.3, 99.6, 99.6, 99.6

Previous work (4, 5) has shown that tropic acid, the acid moiety of the natural solanaceous alkaloids, is not oxidized by ceric salts, therefore, the test for *Atropine and Other Solanaceous Alkaloids* in the USP XVII monograph would be unnecessary if cerimetric titration were used for the assay of homatropine. The selectivity inherent in the assay method would also eliminate the need for the test for *Most Other Alkaloids* in the homatropine hydrobromide monograph.

Use in Stability Evaluations—The kinetic study reported by Patel and Lemberger (2) on homatropine hydrolysis demonstrated that solutions of the drug have limited stability at the pH range recommended in USP XVII. Thus, the availability of a convenient method for stability evaluation of homatropine solutions is of more than theoretical interest in assessing product potency.

The general rate equation (2) affords a calculated first-order rate constant of 6.6×10^{-3} min.$^{-1}$ for the hydrolysis of homatropine at pH 9.95 and 30°. The average of four duplicate samples taken over a period of about 100 min. yielded an estimate of 6.0×10^{-3} min.$^{-1}$ for this constant.

SUMMARY AND CONCLUSIONS

Cerimetric titration of mandelic acid liberated by the saponification of homatropine is proposed as an assay for homatropine hydrobromide and homatropine hydrobromide ophthalmic solution. The titration is conducted on a chilled solution to circumvent interference of the tropine component of the saponification products, for tropine is slowly oxidized at room temperature. The selectivity of the oxidation reaction for arylglycolic acids eliminates the need for the tests for *Atropine and Other Solanaceous Alkaloids* and *Most Other Alkaloids* in the USP XVII monograph for homatropine hydrobromide. The proposed assay has the important advantage in comparison with the USP XVII procedure of being stability indicating. Like the official assay, the proposed method is absolute, *i.e.*, it does not require use of a reference standard.

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Keyphrases

Homatropine HBr—stability determination
Cerimetric titration, direct—analysis
Hydrolysis, homatropine HBr—detection

Technical Articles

Study on Dosage Variations of Individual Capsules and Tablets of Desipramine and Imipramine Hydrochloride

By S. AHUJA, C. SPITZER, and F. R. BROFAZI

Automated methods of analyses based on UV absorption of desipramine and imipramine hydrochloride were developed in order to obtain information on intercapsule and intertablet variations of the following six dosage forms: desipramine hydrochloride capsules 10, 25, and 50 mg. and imipramine hydrochloride tablets 10, 25, and 50 mg. The automated methods require the use of an automatic analyzer and the same manifold is used for all six dosage forms. A careful review of the results obtained on individual capsules and tablets reveals that almost all of the capsules and tablets analyzed by these methods were within ±15 percent limits of the indicated dosage.

THE PHARMACEUTICAL INDUSTRY has long been interested in obtaining more information on drug content of individual tablets or capsules to assure high standard of drugs, from the standpoint of production, quality control, and therapeutic activity. This information is necessary for establishing suitable control measures in production and quality control in order to obtain reasonable dosage uniformity of drugs.

Analytical methods which are based on the analysis of sample composites cannot provide reliable indications of content uniformity because they express product dosage on individual dosage forms, whereas the analyses are actually run on sample composites. Therefore, such analyses not only average out small variations in composition between individual tablets or capsules, but may also mask large deviations.

Quantitative analysis of the active ingredient itself, in each of the individual capsules or tablets, should provide a good approach for studying dosage variations. However, this approach was rather difficult to carry out until the recent introduction of automated methods of analyses.

The automated methods of analyses based on UV absorption of desipramine hydrochloride[1] [10,11 - dihydro - 5 - (3-methylaminopropyl) - 5*H*-dibenz[*b*,*f*] azepine hydrochloride] and imipramine hydrochloride[2] [10,11 - dihydro - 5 - (3-dimethylaminopropyl) - 5*H*-dibenz[*b*,*f*]azepine hydrochloride] were developed with the object of gaining information on intercapsule and intertablet variations on the following six dosage forms: (*a*) desipramine hydrochloride capsules (hard gelatin) 10, 25, and 50 mg., and (*b*) imipramine hydrochloride tablets (sugar coated) 10, 25, and 50 mg.

A variety of methods (1–8) have been used for the analysis of desipramine and imipramine hydrochloride. For several years manual UV methods have been used for the analysis of these compounds in these laboratories. These pro-

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[1] Pertofrane, Geigy Chemical Corp., Ardsley, N. Y.
[2] Tofranil, Geigy Chemical Corp., Ardsley, N. Y.