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Ion-Exchange Separation and Ultraviolet Determination of Phenylephrine, Codeine, and Selected Antihistamines

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A method for the isolation and determination of phenylephrine, codeine, or an antihistamine as a single active ingredient is presented. Also, a method is presented for the separation and determination of an antihistamine and codeine or an antihistamine and phenylephrine in pharmaceutical products containing these combinations. All of the amines are extracted with a strong cation-exchange resin, AG 50W-X4. The phenylephrine or codeine is eluted with 1 *N* hydrochloric acid in 60 per cent methanol in water. The antihistamine is then eluted with 3.5 *N* hydrochloric acid in 40 per cent methanol in water. The compounds are determined by subjecting the eluates to ultraviolet spectrophotometry. The assay is used successfully on several commercial products.

A LARGE NUMBER of pharmaceuticals are presently marketed for treatment of the common cold and other respiratory disorders. These products use a large variety of active ingredients but for the most part they are, chemically, basic amines. These amines are in the form of vasoconstrictors, antitussives, and antihistamines.

Phenylephrine is widely used as a vasoconstrictor in these products. The assay for this compound in liquid pharmaceuticals has been difficult because of the water solubility of phenylephrine even as the base. The U.S.P. (1) describes the time honored bromination procedure. This procedure is ineffective when oxidizable substances are present because of their reactions with bromine. Reducing sugars are a good example of this type of interference. The reactions of phenols with ferricyanide and 4-aminoantipyrine has been proposed by several workers (2, 3). Chafetz (4) oxidized phenylephrine and other phenylethanolamines to their aromatic aldehydes, extracted with organic solvents, and determined the ultraviolet absorption. Clark and Rosenberg (5) reported phenylephrine separation using a Levine column with acetylation on the column. The phenylephrine was determined in the eluates. Auerbach (6) used a diazotization technique where phenylephrine acted as the coupler. Kelly and Auerbach (7) utilized ion-exchange chromatography in a method similar to that presented in this paper.

Codeine, which is a very popular antitussive, has been the subject of extensive analytical study for many years because of its wide usage. Nonaqueous titrimetry has been utilized by several workers (8–10). Artamonov and Bugreeva (11) applied the techniques of high frequency titration for the determination of codeine.

Several colorimetric methods have been proposed (12, 13) as well as fluorimetry (14, 15), refractometry (16), and spectrophotometry (17, 18).

Various types of chromatographic procedures have been used for codeine analysis. Izmailov and co-workers (19) used paper chromatographic techniques to separate codeine from other poppy extract alkaloids. Adsorption columns of silicic acid were utilized by Andreeva and Figurovskii (20). The more recent techniques of gas chromatography (21) and thin-layer chromatography (22) have also been reported.

The common methods of assay for antihistamines include nonaqueous titrimetry (23, 24), ultraviolet spectrophotometry (25), and reineckate salt formation with subsequent colorimetric determination of the acetone solution (26). A widely used colorimetric method, specific for compounds containing the 2-pyridyl group, is based on the König reaction with cyanogen bromide and aniline. This was described by Jones and Brody (27) and later modified by Hudanick (28). Gas chromatography has been used successfully by Celeste and Turczan (29) and by Fontan and co-workers (30).

This paper utilizes strongly acidic cation-exchange resins to separate the amines from common dosage form ingredients. The amines are then eluted separately from the resin using specific concentrations of hydrochloric acid. The amines are deter-

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mined in the eluate by ultraviolet spectrophotometry.

EXPERIMENTAL

Apparatus.—Glass column 20 cm. × 1 cm. with stopcock made of Teflon and containing built in needle valve for control of flow rate. The column is also fitted with a reservoir with a capacity of 250 ml.

A suitable recording ultraviolet spectrophotometer such as the Beckman DK-2A or Spectronic 505 which records in absorbance units.

Reagents.—Cationic exchange resin AG 50W-X4 100–200 mesh in hydrogen form available from Bio-Rad Laboratories, Richmond, Calif. Enough resin, about 3 Gm., is added in the form of a slurry to the glass column and rinsed with water.

Hydrochloric acid, 0.05 *N* in 50% methanol in water. Hydrochloric acid, 1.0 *N* in 60% methanol in water. Hydrochloric acid, 3.5 *N* in 40% methanol in water.

Standard Solutions.—Prepare the following standard solutions using U.S.P., N.F., or other suitable reference standards.

(*a*) Phenylephrine hydrochloride, 10 mg./200 ml., 1.0 *N* hydrochloric acid in 60% methanol in water. (*b*) Codeine phosphate, 20 mg./200 ml., 1.0 *N* hydrochloric acid in 60% methanol in water. (*c*) Chlorpheniramine maleate, 4 mg./200 ml., 3.5 *N* hydrochloric acid in 40% methanol in water. (*d*) Pheniramine maleate, 6 mg./200 ml., 3.5 *N* hydrochloric acid in 40% methanol in water. (*e*) Promethazine hydrochloride, 12.5 mg./200 ml., 3.5 *N* hydrochloric acid in 40% methanol in water. (*f*) Methapyrilene hydrochloride, 4 mg./200 ml., 3.5 *N* hydrochloric acid in 40% methanol in water.

Sample Treatment.—Use 10-ml. samples of each product except antihistamines *A*[1] (5 ml.), *B*[2] (20 ml.), and *C*[3] (20 ml.). Pipet the sample into the reservoir, rinse the pipet with distilled water, and add to the reservoir.

Add distilled water to the sample to make the volume approximately 100 ml. and mix well.

Allow the sample solution to flow through the resin bed at the rate of 2–3 ml./min. Wash the column by adding 100 ml. of distilled water and allow it to flow through the resin at the rate of 5 ml./min.

Traces of aromatic amines from flavors or coloring agents are removed by allowing 50 ml. of 0.05 *N* hydrochloric acid in 50% methanol in water to flow through the column at 5 ml./min.

For products containing phenylephrine or codeine as a single active ingredient, place a 200-ml. volumetric flask under the column and add 190 ml. of 1.0 *N* hydrochloric acid in 60% methanol in water to the reservoir. Allow this to flow through the column at the rate of 3 ml./min. The volume is adjusted with 1.0 *N* hydrochloric acid in 60% methanol in water.

For products containing an antihistamine as a single amine active ingredient, position a 200-ml. volumetic flask under the column and add 190 ml. of 3.5 *N* hydrochloric acid in 40% methanol in water to the reservoir. Allow this to flow through the column at the rate of 4 ml./min. The volume is adjusted with 3.5 *N* hydrochloric acid in 40% methanol in water.

For products containing either phenylephrine or codeine in addition to an antihistamine, follow the same procedures outlined above eluting the phenylephrine or codeine first with 1.0 *N* hydrochloric acid in 60% methanol in water, then eluting the antihistamine from the same column using 3.5 *N* hydrochloric acid in 40% methanol in water.

Determination.—Quantitatively dilute the sample eluates, if necessary, so that the theoretical concentrations are as close as possible to the standard concentrations. Record the ultraviolet spectra of the final sample dilutions and the standard solutions using a suitable spectrophotometer. Absorbance units should be used.

Using the base line technique, determine the absorbance for each sample and standard at the particular maximum. Calculate the amounts of each compound present in each sample from the values obtained for the standard solutions.

DISCUSSION AND RESULTS

Standard solutions, when subjected to the above procedures, gave the results shown in Table I. These results are derived from 10 determinations for each compound. These data indicate that the method is accurate to within 1% of the true values and are reproducible to ±1.5% at 95% confidence limits. The data for the analyses of marketed products are shown in Tables II and III.

A slightly raised base line was noted with some products where a small portion of the coloring agent was held and eluted with the sample. This did not cause any problem since the base line technique was used in the calculations.

When the maleate salt of a compound is used for a standard, there is a small difference in the base line caused by ultraviolet absorbance of the maleic acid

Table I.—Accuracy, Reproducibility, and Precision Analysis Data

Std. Material	Concn.	Means ($\bar{X}$) of Results of 10 Determinations in % of Theoretical	S.D. of 10 Determinations in % of Theoretical
Phenylephrine HCl	20 mg./10 ml.	99.4	±0.61
Codeine phosphate	20 mg./10 ml.	99.9	±0.64
Chlorpheniramine maleate	4 mg./10 ml.	100.4	±0.44

Table II.—Analysis of Products Containing a Single Amine Component

Ingredient	Label Claim	Found % of Label Claim
Phenylephrine HCl[a]	5 mg./5 ml.	101.2
Codeine phosphate[b]	1 Gm./fl. oz.	102.5
Codeine[c]	0.91 Gm./fl. oz.	98.5
Promethazine HCl[d]	6.25 mg./5 ml.	98.1
Methapyrilene HCl[e]	4 mg./ml.	98.9

[1] Marketed as Histadyl Syrup by Eli Lilly & Co., Indianapolis, Ind.

[2] Marketed as Demazine Syrup by Schering Corp., Union, N. J.

[3] Marketed as Novahistine Elixir by Pitman-Moore, Indianapolis, Ind.

[a] Neo-Synephrine Elixir, Winthrop Laboratories, New York, N. Y. [b] Cheracol, The Upjohn Co., Kalamazoo, Mich. [c] Elixir Terpin Hydrate and Codeine, Parke-Davis and Co., Detroit, Mich. [d] Phenergan Syrup, Wyeth Laboratories, Philadelphia, Pa. [e] Histadyl Syrup, Eli Lilly and Co., Indianapolis, Ind.

TABLE III.—ANALYSIS OF PRODUCTS CONTAINING TWO AMINE COMPONENTS

Ingredients	Label Claim	Found % of Label Claim
Codeine phosphate[a]	10 mg./5 ml.	98.7
Pheniramine maleate[a]	7.5 mg./5 ml.	103.3
Phenylephrine HCl[b]	2.5 mg./5 ml.	98.5
Chlorpheniramine maleate[b]	1.25 mg./5 ml.	100.3
Phenylephrine HCl[b]	5 mg./5 ml.	99.4
Chlorpheniramine maleate[c]	1 mg./5 ml.	100.2

[a] Robitussin AC, A. H. Robins Co., Richmond, Va. [b] Demazine Syrup, Schering Corp., Union, N. J. [c] Novahistine Elixir, Pitman-Moore Co., Indianapolis, Ind.

radical. This also is overcome for the most part by the use of the base line technique.

One deviation from the described procedures was necessary for antihistamine *D*[4]. Since terpin hydrate is precipitated on dilution with water, the sample dilution prior to column application and the washing of the column were performed using 50% methanol in water instead of distilled water.

The proposed method was successful in the presence of normal syrup and elixir components in addition to other materials such as menthol, glyceryl guaiacolate, terpin hydrate, ammonium chloride, tartar emetic, potassium guaiacolsulfonate, and chloroform.

SUMMARY

A method of assay for liquid dosage forms containing phenylephrine, codeine, or an antihistamine has been presented. Also included was an assay method for combinations of phenylephrine or codeine with an antihistamine. The procedures have been used successfully on commonly available products with accurate and reproducible results.

[4] Marketed as Elixir Terpin Hydrate with Codeine by Parke-Davis & Co., Detroit Mich.

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