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Direct Determination of Quinine Ethylcarbonate with Monoprotic Acid Dye by Solvent Extraction

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A singly charged tetrabromophenolphthalein ethyl ester (TBPE) was found to be extracted with quinine ethylcarbonate as a 1:1 complex in 1,2-dichloroethane. Among various solvents, TBPE-1,2-dichloroethane system gives the red colored species for alkaloids and amines, while in the absence of alkaloids and amines, the organic phase shows pale yellow color. In this way, the spectrophotometric method was investigated for the determination of a small amount of quinine ethylcarbonate by solvent extraction. Quinine ethylcarbonate is determined by measuring absorbance of the extracts over the range of 2×10^{-6} M to 1.6×10^{-5} M (0.793—6.34 μg/ml) at 555 nm. From the electric conductivity measurements, the extracts with TBPE are classified into four categories as follows; (1) blue; dissociated ion-pair complexes (2) blue; associated ion-pair compounds (3) red; associated charge transfer complexes (4) yellow; associated H·TBPE.

Introduction

A number of quaternary ammonium salts, amines and alkaloids are extracted quantitatively into 1,2-dichloroethane with tetrabromophenolphthalein ethyl ester (TBPE) which is a singly charged dye and the extraction reactions have been applied to the photometric determination of some quaternary ammonium salts,[2] amines[3] and alkaloids.[4]

This paper deals mainly with the spectrophotometric determination of quinine ethylcarbonate as a series of investigations with TBPE. The advantages of the proposed method are a high sensitivity and a wide optimum pH range to extract in comparison with other methods using diprotic acid dyes, such as bromothymol blue[5] and bromophenol blue.[6] In addition, quinine ethylcarbonate can be determined directly without hydrolysis by using this method. Moreover, in the course of these investigations, the extracts were found to be classified into the following four categories from the electric conductivity; (1) blue; dissociated ion-pair complexes (2) blue; associated ion-pair compounds (3) red; associated charge transfer complexes (4) yellow; associated H·TBPE.

The titrimetric[7] and spectrophotometric[8] methods have been used for the determination of quinine ethylcarbonate. However, those methods are not favourable in the analysis of pharmaceutical preparations because these suffer from interference of many substances and the procedure is somewhat complicated. Recently, bromphenol blue[9] was proposed for the spectrophotometric determination of quinine ethylcarbonate by solvent extraction. However, the method is an indirect determination of quinine ethylcarbonate by hydrolysis.

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Experimental

Apparatus——(1) The spectrophotometric measurements were made with a Hitachi Model 124 spectrophotometer with 10 mm cells.

(2) The pH measurements were made with a Hitachi-Horiba Model M-5 pH meter.

(3) An Iwaki Model KM shaker was used for the extraction.

(4) A Toa Denpa Model CM-2A electric conduct meter was used for the measurements of electric conductivity.

Reagents——TBPE Solution: The solution of 4.0×10^{-3} M was prepared by dissolving 0.2800 g of TBPE potassium salt (mol. wt. 700.1) in ethyl alcohol and diluting the solution to 100 ml with ethyl alcohol.

Standard Quinine Ethylcarbonate Solution: The stock solution of 1.0×10^{-2} M was prepared by dissolving 3.965 g of quinine ethylcarbonate in dilute hydrochloric acid and diluting the solution to 1 liter with water. The pH 8.5 buffer solution was prepared by mixing a 0.4 M potassium dihydrogen phosphate solution containing 0.08 M sodium borate and a 3 N sodium hydroxide solution.

All the chemicals were of reagent grade, and distilled water was used.

Recommended Procedure——Take 0.5—4 ml of the standard quinine ethylcarbonate solution (1×10^{-4} M), 2 ml of TBPE and 5 ml of the buffer solution (pH 8.5) into a 100 ml separatory funnel. Dilute the mixture to 25 ml with water and shake the solution for 3 min with 10 ml of 1,2-dichloroethane. After the separation of the two layers, run off the extract into a glass tube through a filter paper to remove droplets of water. Measure the absorbance of the extract at 555 nm, using a reagent blank or water as a reference.

A linear relationship between the absorbance of the extract and the concentration of the sample in the aqueous solution was observed for the concentration range 2×10^{-6} M to 1.6×10^{-5} M (0.793—6.34 μg/ml) of quinine ethylcarbonate in the aqueous solution. The absorbance for 8×10^{-6} M quinine ethylcarbonate is 0.510 at 555 nm. The relative standard deviation was about 1%, which was estimated from the results of ten sample solutions for 8×10^{-6} M quinine ethylcarbonate.

Results and Discussion

Absorption Spectra

In the absence of quinine ethylcarbonate a yellow compound is extracted into 1,2-dichloroethane, whilst in the presence a red one (λ_{max} 555 nm) is formed. The extract may be attributed to the formation of an addition compound between TBPE and quinine ethylcarbonate.

Effect of pH

The effect of pH on the extraction was studied by extracting quinine ethylcarbonate together with TBPE from a series of aqueous solutions buffered at various pH values. The maximum and constant absorbance of the extract was obtained when the pH of the aqueous phase was in the range of 8.0 to 9.5.

Inoue[9] has reported the indirect determination of quinine ethylcarbonate with the bromophenol blue anion, which is diprotic acid dye. This method is based on the extraction of quinine with bromophenol blue after the hydrolysis of quinine ethylcarbonate by adding sodium hydroxide. Moreover, the bromophenol blue anion is used as a singly charged anion by adjusting the pH at 6.2. Consequently, quinine ethylcarbonate may be extracted as $[Quinine]^+\cdot[Bromophenol\ blue]^-$ ion-pair. This is perhaps because the ion-pair formation is restricted owing to the following factors; (1) the presence of the bulky ester group near the positive charge (2) the large size of the sulfonic group (3) the delocalized charge on the sulfonic group. In the case of the proposed method, however, it is possible to determine quinine ethylcarbonate directly. Because of the higher charge density of the TBPE anion, an associated charge transfer complex may be formed between quinine ethylcarbonate and TBPE and extracted into 1,2-dichloroethane. Moreover, TBPE is more hydrophobic than bromophenol blue. Chart 1 shows the extraction mechanism of the quinine ethylcarbonate cation with TBPE anion.

The Other Variables

It was found that the concentration of TBPE should be maintained at more than 30 fold molar excess over quinine ethylcarbonate (8×10^{-6}M in aqueous phase) to obtain a maximum and constant extraction. Excess amounts of the phosphate buffer had no influence on the

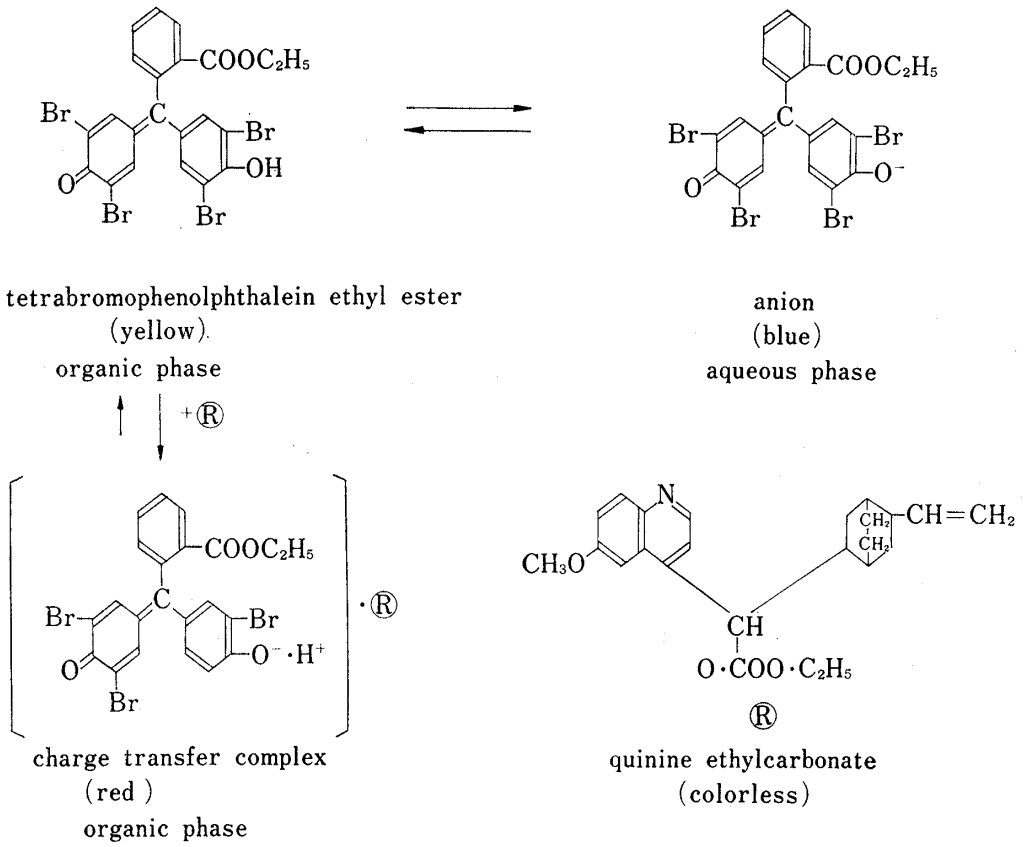

Chart 1

absorbance of the extract. Full color development took place by shaking for about 1 min. The color intensity of 1,2-dichloroethane extracts remained constant for at least 1 hour. The fluctuations of room temperature gave no appreciable effect on the absorbance.

Effect of Foreign Substances

Chloride, bromide, magnesium, glucose and starch which are apt to exist in pharmaceutical preparations with quinine ethylcarbonate do not interfere. While a small amount of alkaloids, such as papaverine, caffeine or quaternary ammonium, such as benzethonium, gave strong interferences.

Table I. Electric Conductivity of Extracts

Aqueous phase (50 ml) containing	Color of extracts	Electric conductivity in solvents ($\mu\nu$/cm)	
		Dichloroethane	Toluene
10^{-4}M TBPE[a] only	yellow	0.0	0.0
10^{-4}M berberine and 10^{-4}M TBPE	blue	3.9	0.0
10^{-4}M quinine ethylcarbonate and 10^{-4}M TBPE	red	0.0	0.0
10^{-4}M sparteine and 10^{-4}M TBPE	blue (in dichloroethane)	3.8	—
	red (in toluene)	—	0.0

a) TBPE: tetrabromophenolphthalein ethyl ester
b) Extract with 40 ml of 1,2-dichloroethane or toluene was filetred with a filter paper to remove droplets of water, and the conductivity was measured at 25°.
c) The pH was kept at 8.5 with the phosphate buffer.

Investigation of the Extracted Species

In the previous paper,[10] the colors of the extracts into 1,2-dichloroethane were classified into three categories; (1) blue extracts which are observed in the presence of the quaternary ammonium salts (2) red extracts which are developed in the presence of alkaloids (3) yellow extracts which are the same color as the reagent blank. In this paper, the extracts into 1,2-dichloroethane or toluene are classified into four categories by electric conductivity in Table I; (1) blue; dissociated ion-pair complexes, *e.g.*, berberine, benzethonium, sparteine in 1,2-dichloroethane (2) blue; associated ion-pair compounds, *e.g.*, berberine in toluene (3) red; associated charge transfer complexes, *e.g.*, ephedrine, quinine ethylcarbonate, procaine in 1,2-dichloroethane or sparteine in toluene (4) yellow; associated H·TBPE. These phenomena can be attributed to the complicated interactions between the cation and TBPE which are sensitively influenced by the molecular structure of the cation and the dielectric nature of solvents. Consequently, the extracted species may be formulated as [quinine ethylcarbonate]·H·[TBPE] from continuous variation plots.

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Studies on the Constituents of *Mallotus japonicus* Muell. Arg. II.[1] A Corotoxigenin Trioside from the Seeds[1]

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A new cardiac trioside was isolated from the seeds of *Mallotus japonicus* Muell. Arg. (Euphorbiaceae) and it was identified as corotoxigenin β-D-glucopyranosyl-(1→6)-β-D-glucopyranosyl-(1→4)-α-L-rhamnopyranoside.

In the preceding paper, the authors reported isolation and characterization of α-L-rhamnopyranosides and β-D-glucopyranosyl-(1→4)-α-L-rhamnopyranosides of corotoxigenin, mallogenin, coroglaucigenin and panogenin, aglycones common to those in *Mallotus philippinensis* and *M. paniculatus*.[3] The authors mentioned that the glucorhamnosides were the genuine cardiac glycosides. However, one more polar compound sensitive to the Kedde reaction was detected on closer examination of the intermediate emulsion layer which formed on butanol extraction of water layer after a series of solvent extraction. This paper deals with characterization of this newly detected cardiac glycoside (I).

The emulsion layer was separated and evaporated to a dark brown powder and the aqueous solution was passed through the polyamide column. The eluate was colorless but I could not be separated from other polar contaminants. Silica gel column chromatography of the

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