THE PHOTOMETRIC DETERMINATION OF CONDELPHINE

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Condelphine, an alkaloid found in certain species of delphinium [1-4], causes a curare-like effect and is applied clinically for nervous disorders with a pathologically increased muscular tone, and also for curing tetanus [5-7].

The existing method for the quantitative determination of condelphine is based on the calculation of the content of acetic acid formed in the saponification of the alkaloid. This method is laborious and requires a large amount of material.

We propose a method which makes it possible to carry out the quantitative determination rapidly, with high accuracy, and a minimum loss of material.

It is known that organic bases and acidic dyes form colored compounds which under certain conditions are extracted with organic solvents. We were guided by the data of A. K. Babko and V. S. Konyushko on the extraction-photometric determination of alkaloids [8,9]. They used bromphenol blue (BPhB) as the reagent. Here it was established that upon mixing aqueous solutions of condelphine and BPhB, a colored compound is formed which is extracted with chloroform. Since the pH has a significant effect on the extraction of the colored compound [9], we preliminarily studied the effect of pH on the formation and extraction of the colored compound.

The work was carried out in the following way. To 2 ml of a solution of condelphine (5 \times 10^{-4} \text{ M solution of alkaloid in 0.01 N solution of HCl}) was added 2 ml of a solution of BPhB (5 \times 10^{-4} \text{ M solution of BPhB in 0.01 N solution of NaOH}) and the pH was brought to the required value using buffered solutions. The formed product was extracted with 10 ml of chloroform and the optical density of the chloroform extract was measured in a FÉK-56 photoelectrocalorimeter having a blue light filter (\lambda = 440 nm [9]). The obtained data are presented in Fig. 1. The maximum optical density is reached at pH 2.6 - 3.6. It should be noted that the optimum pH of extraction corresponds completely to the theoretical pH determined by the equation [9] pH = 14 - pKa. It is known that the pKa of condelphine is equal to 7.6 [11].

The stoichiometric ratio of the reacting compounds was established by the method of isomolar series [12]. The graph of the isomolar series was constructed in the following way. Isomolar solutions of condelphine and BPhB were mixed in certain ratios; the pH was brought to 3.2 with a biphthalic buffer solution [10], and the formed product was extracted with 10 ml of chloroform. The optical density was measured on a FÉK-56 photoelectrocalorimeter. From the obtained data was constructed a graph of the dependence of the optical density on composition (Fig. 2, A). As is seen from the obtained data, the composition of the reaction product corresponds to a molar ratio of the condelphine and BPhB components of (2:1), which agrees with literature data [9]. The constant of formation of the colored compound, calculated by us from the known equation [13], is equal to (1.1 \pm 0.1) \times 10^{-7}. The coefficient of the molar extinction of the formed compound is 25,600. The specific extinction coefficient calculated for condelphine is 286.

A method for the quantitative determination of condelphine was developed based on the studied reaction of the interaction of condelphine with BPhB. The determinations were carried out using the calibration graph which was constructed in the following way. Into a separatory
funnel were placed 0.5-3 ml of a 0.015% solution of condelphine and in 5 ml quantities, a $5 \cdot 10^{-4} \text{M}$ solution of BPhB. The pH was brought to 3.2 by addition of a biphthalie buffer solution. The formed compound of BPhB with condelphine was extracted with 10 ml of chloroform and the optical density was measured with a blue light filter (Fig. 2, B).

The error of the method was established in the following way. A weighed portion of compound was dissolved in a 0.01 N solution of HC1 and the solution was brought to 100 ml. The content of the preparation was determined from the calibration graph. One ml portions of the obtained solution of condelphine in powders and their treatment are presented in Table 1.

\[
S_x = \sqrt{\frac{\sum (x_i - \bar{x})^2}{n(n-1)}} = \sqrt{\frac{10}{7(7-1)}} = 0.5 \quad \text{(average quadratic deviation from the average arithmetic)}.
\]

\[
\varepsilon = S_x \cdot t_{a} \cdot K = 0.5 \cdot 2.447 = 1.22 \quad \text{(absolute error)};
\]

\[
\varepsilon_{rel} = \frac{\text{rel} \cdot 100}{0.001 \cdot 100} = 1.22\% \quad \text{(relative error)}.
\]

As is seen from the presented data, the relative error of the method does not exceed $\pm 1.3\%$.

\[
S_x = \sqrt{\frac{4.62 \cdot 10^{-4}}{30}} = 0.39 \cdot 10^{-3},
\]

\[
\varepsilon = 0.39 \cdot 10^{-3} \cdot 2.570 = 1.0 \cdot 10^{-8},
\]

\[
\varepsilon_{rel} = \frac{0.001 \cdot 100}{0.025} = 4\%.
\]

In the determination of condelphine in tablets, the tablet of the preparation containing 0.025 g of condelphine was powdered in a mortar, dissolved in a 0.01 N solution of HC1, and filtered into a 100 ml meas-

### TABLE 1. Results of the Quantitative Determination of Condelphine in the Pure Preparation

<table>
<thead>
<tr>
<th>Condelphine taken (in g)</th>
<th>Optical density</th>
<th>Found</th>
<th>(x_i - \bar{x})</th>
<th>((x_i - \bar{x})^2)</th>
</tr>
</thead>
<tbody>
<tr>
<td>0.0131</td>
<td>0.370</td>
<td>0.0130</td>
<td>99.2</td>
<td>-0.9</td>
</tr>
<tr>
<td>0.0110</td>
<td>0.320</td>
<td>0.0112</td>
<td>101.8</td>
<td>+1.7</td>
</tr>
<tr>
<td>0.0144</td>
<td>0.465</td>
<td>0.0163</td>
<td>99.4</td>
<td>-0.6</td>
</tr>
<tr>
<td>0.0210</td>
<td>0.606</td>
<td>0.0212</td>
<td>101.0</td>
<td>+0.9</td>
</tr>
<tr>
<td>0.0246</td>
<td>0.700</td>
<td>0.0245</td>
<td>99.0</td>
<td>-1.1</td>
</tr>
<tr>
<td>0.0315</td>
<td>0.890</td>
<td>0.0311</td>
<td>98.7</td>
<td>-1.4</td>
</tr>
<tr>
<td>0.0319</td>
<td>0.526</td>
<td>0.0324</td>
<td>101.5</td>
<td>+1.4</td>
</tr>
</tbody>
</table>

\[
\Sigma = 700.6 \quad \Sigma = 10.00
\]

### TABLE 2. Results of the Determination of Condelphine in Tablets

<table>
<thead>
<tr>
<th>By prescription (in g)</th>
<th>Found (in g)</th>
<th>(x_i - \bar{x})</th>
<th>((x_i - \bar{x})^2)</th>
</tr>
</thead>
<tbody>
<tr>
<td>0.0225-0.0275</td>
<td>0.0250</td>
<td>+0.0010</td>
<td>1.00</td>
</tr>
<tr>
<td>0.0225-0.0275</td>
<td>0.0240</td>
<td>-0.0010</td>
<td>1.00</td>
</tr>
<tr>
<td>0.0225-0.0275</td>
<td>0.0245</td>
<td>-0.0004</td>
<td>0.18</td>
</tr>
<tr>
<td>0.0225-0.0275</td>
<td>0.0239</td>
<td>-0.0011</td>
<td>1.21</td>
</tr>
<tr>
<td>0.0225-0.0275</td>
<td>0.0260</td>
<td>+0.0010</td>
<td>1.00</td>
</tr>
<tr>
<td>0.0225-0.0275</td>
<td>0.0255</td>
<td>+0.0005</td>
<td>0.25</td>
</tr>
</tbody>
</table>

\[
\Sigma = 0.1500 \quad \Sigma = 4.62
\]

\[
\chi = 0.025
\]