Kinetic spectrophotometric determination of Co(II) ion by the oxidation of Ponceau 4R by hydrogen peroxide

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Abstract: A new, sensitive and simple kinetic method has been developed for the determination of traces of Co(II) ions based on their catalytic effect in the oxidation of trisodium-2-hydroxy-1-(4-sulphonato-1-naphthylazo)naphthalene-6,8-disulphonate (red artificial color Ponceau 4R) by hydrogen peroxide in borate buffer. The reaction was followed spectrophotometrically by tracing the oxidation product at 478.4 nm within 1 min after the initiation of the reaction. The optimum reaction conditions are: borate buffer (pH 10.5), Ponceau 4R (8 × 10⁻⁶ mol/dm³), H₂O₂ (3 × 10⁻² mol/dm³) at 22 °C. Following this procedure, Co(II) can be determined with a linear calibration graph up to 1.17 ng/cm³ and a detection limit of 0.20, based on the 3σ criterion. The relative error ranges between 4.80–3.25 % for the concentration interval of Co(II) ions 1.76–17.61 ng/cm³. The effects of certain foreign ions on the reaction rate were determined for an assessment of the selectivity of the method. The method was applied for the determination of Co(II) in pharmaceutical samples.

Keywords: Co(II) determination, kinetic spectrophotometric method, Ponceau 4R oxidation, pharmaceutical samples.

INTRODUCTION

Cobalt is an essential element for the functioning of many vital processes. It is extremely important in the processes of blood formation, stimulation of hemoglobin synthesis, and the functioning of vitamins, enzymes and hormones. This metal has a very positive influence on the metabolism of vitamins, such as ascorbic acid and vitamin B₁₂. Cobalt is an essential element in the human body as a component of vitamin B₁₂ and it is clear that the monitoring of body fluids for cobalt is important for the control of nutritional deficiencies and, perhaps, the prevention of its toxic effect in cases of occupational exposure. Finally, it is also necessary for the synthesis of a number of hormones, neurotransmitters, and other compounds, such as bile acids and DNA.¹²

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The human organism is supplied with cobalt through food and water, therefore it is necessary to determine and control its concentration in both.

In recent years, kinetic methods have become important in trace analysis and the application of catalytic reactions in trace analysis has been reviewed previously. Many methods for the kinetic determination of Co(II) are based on its catalytic effects in redox reactions, including ligand-substitution reactions, or in the oxidation of organic compounds by H₂O₂. The oxidation of these compounds leads to the formation of colored products, hence the kinetics of the trace reactions are usually followed spectrophotometrically. Most of these methods are very sensitive in the range of nanograms, and do not demand costly equipment.³⁻¹¹ Among these methods, electrothermal atomic absorption spectrometric methods have also been used for the determination of Co(II).¹² This procedure permits the determination of 0.02 μg/dm³ Co(II) in serum samples with a relative standard deviation of 10–18 %.

Flame atomic absorption spectrometry has been used for Co(II) determination. The lowest concentration for quantitative recovery was 4 ng/cm³ of Co(II). The method was used for Co(II) determination in pharmaceutical samples.¹³ Flow-injection analysis has also been used for Co(II) determination with a high sensitivity and selectivity, but the methods suffer, from more or less time consuming procedures and/or expensive and complicated instrumentation.¹⁴,¹⁵

EXPERIMENTAL

Apparatus
A spectrophotometric method was used for following the rate of the investigated reaction. The dependence of the absorbance (A) on (t) was measured using a Perkin-Elmer Lambda 15 spectrophotometer, connected to a thermocirculating bath. The pH was measured by means of Radiometer PHM 29b pH meter and a combined glass–calomel electrode, GK 2311C. The solution was thermostated at 22 ± 0.1 ºC before the beginning of the reaction.

Reagents
A hydrogen peroxide solution was prepared from 30 % reagent (c = 1 mol/dm³) by diluting with deionized water.

A solution of the artificial color Ponceau 4R (c = 1 × 10⁻³ mol/dm³) was prepared by dissolving it in deionized water. The working Ponceau 4R solution (c = 1.6 × 10⁻⁴ mol/dm³) was obtained by diluting with water.

The borate buffer solution was prepared by mixing Na₃B₄O₇ (c = 0.05 mol/dm³) with NaOH (c = 0.1 mol/dm³).¹⁶

A solution of CoCl₂ was prepared by dissolving CoCl₂ in water (c = 1 × 10⁻⁵ mol/dm³) which had been standardized gravimetrically.

All chemicals were of analytical reagent grade and were provided by Merck unless indicated otherwise. The artificial color Ponceau 4R was obtained from food industry “HISAR”. The solutions were prepared using deionized water. All the stock solutions were stored in polyethylene containers. All the polyethylene containers and the glassware used were cleaned in aqueous HCl (1:1) and then thoroughly rinsed with deionized water.

Procedure
The selected volumes of reagents were transferred into a 10 cm³ volumetric flask (Boudierin flask) in the order borate buffer, Ponceau 4R, hydrogen peroxide and catalyst. Water was added to
give the predetermined volume. The flask was thermostated for 10 min, and vigorously shaken. The spectrophotometer cell was rinsed well and filled with the solution. The absorbance at 478.4 nm, was measured every 30 s over a period of 4 to 6 min after mixing. Instead of the reaction rate \( \frac{dc}{dt} \), the quantity \( \frac{dA}{dt} \) was used. The measurements were performed at 22 ± 0.1 °C.

RESULTS AND DISCUSSION

Kinetic studies

An integral variant of the tangent method was used for processing of the kinetic data, because a linear correlation exists between the logarithm of the absorbance and time during the first 4 to 6 min after mixing.\(^{17}\) The reaction rate was followed by the change in the values of the tangent of the angle (\( \tan \alpha \)) of the slope of the linear part of the kinetic curve to the abscissa in the coordinates \( \log A – t \), because \( \tan \alpha = \frac{d \log A}{dt} \). Examples of such plots are shown in Fig. 1.

In order to determine the lowest possible determinable concentration of Co(II) ions, the condition needed to be optimized. Therefore, the dependencies of the rates of both the catalytic and the non-catalytic reaction on the concentration of each of the reactants were determined.

Fig. 1. Dependence of the logarithm of the absorbance of the solution on time. Initial conditions: \( c_{H_2O_2} = 3 \times 10^{-2} \text{ mol/dm}^3; c_{\text{Ponceau 4R}} = 8 \times 10^{-6} \text{ mol/dm}^3; \) borate buffer pH 10.50; \( c_{\text{Co(II)}} \) ng/cm\(^3\): 1 – 0.00, 2 – 11.70, 3 – 23.57; \( t = 22 \pm 0.1 \text{ °C} \).

Fig. 2. Dependence of \( \tan \alpha \) on pH for the non-catalytic (1) and catalytic (2) reaction. Initial conditions: \( c_{H_2O_2} = 3 \times 10^{-2} \text{ mol/dm}^3; c_{\text{Ponceau 4R}} = 8 \times 10^{-6} \text{ mol/dm}^3; \) \( c_{\text{Co(II)}} 23.57 \text{ ng/cm}^3; t = 22 \pm 0.1 \text{ °C} \).
The influence of pH on the rate of both reactions is shown in Fig. 2. It can be seen that the greatest difference between the reaction rates occurred at pH 10.50, when Co(II) maximally increases the catalytic reaction rate. For further work, a borate buffer of pH 10.50 was selected.

From Fig. 2 it appears that there is a complicated relationship between the pH and the catalytic reaction rate in the studied concentration range. A linear relationship between the logarithm of $t_g$ and the logarithm of $c_{H^+}$ was found for catalyzed reaction. The order of catalytic reaction is $-0.65$ with respect to $c_{H^+}$ in the investigated interval of $c_{H^+}$ from $3.16 \times 10^{-10}$ to $5.16 \times 10^{-10}$ mol/dm$^3$ (interval pH from 9.50 to 10.50). The non-catalytic reaction is $-1$ order with respect to $c_{H^+}$ in the all investigated interval.

The dependence of the reaction rates on the concentration of H$_2$O$_2$ is shown in Fig. 3, from which it can be seen that the non-catalytic reaction is first order with respect to the H$_2$O$_2$ concentration and catalytic reaction is zero order for the H$_2$O$_2$ concentration from $1 \times 10^{-2}$ to $3 \times 10^{-2}$ mol/dm$^3$, and $-1$ order for higher concentrations. A H$_2$O$_2$ concentration of $3 \times 10^{-3}$ mol/dm$^3$ was selected for further work.

The dependence of the reaction rates on the concentration of Ponceau 4R is shown in Fig. 4, from which it can be seen that the catalytic reaction is first order...
with respect to the Ponceau 4R concentration for all the investigated interval, and the non-catalytic reaction is zero order with respect to the Ponceau 4R concentration.

For further work, a Ponceau 4R concentration of $8 \times 10^{-6}$ mol/dm$^3$ was selected. For the largest studied concentration of $8 \times 10^{-6}$ mol/dm$^3$, the linear part of the kinetic curve is decreasing, and the error of the determination of $t g \alpha$ become larger.

Under the optimal reaction conditions:

pH 10.50 (borate buffer); $c_{H_2O_2} = 3 \times 10^{-2}$ mol/dm$^3$; $c_{\text{Ponceau 4R}} = 8 \times 10^{-6}$ mol/dm$^3$, the Co(II) concentration was varied from 1.17 to 23.57 ng/cm$^3$.

The calibration line, obtained at a temperature of 22 ºC, which can be used for the determination of the Co(II) concentration in the studied interval, is shown in Fig. 5.

The following kinetic equations were deduced on the basis of the graphic correlations obtained for the investigated process.

For the non-catalyzed reaction:

$$-\frac{dc}{dt} = k_0 c_{H^+}^{-1} c_{H_2O_2}$$

were $k_0$ is a constant proportional to the rate constant of the non-catalyzed reaction.

For the catalyzed reaction:

$$-\frac{dc}{dt} = k c_{H^+}^{-0.65} c_{\text{Ponceau 4R}} c(\text{Co(II)})$$

for the $c_{H^+}$ range from $3.16 \times 10^{-10}$ to $5.24 \times 10^{-10}$ mol/dm$^3$ (pH from 9.28 to 10.50) were $k$ is a constant proportional to the rate constant of the catalyzed reaction.

On the basis of these equations, the rate constants for the catalyzed and non-catalyzed reactions were calculated (Table I).

<p>| TABLE I. Rate constants for the non-catalyzed ($k_0$) and catalyzed ($k$) reactions at three temperatures |</p>
<table>
<thead>
<tr>
<th>T/K</th>
<th>$k_0$</th>
<th>$k$</th>
</tr>
</thead>
<tbody>
<tr>
<td>292</td>
<td>$4.60 \times 10^{-7}$</td>
<td>$2.31 \times 10^{-2}$</td>
</tr>
<tr>
<td>295</td>
<td>$6.57 \times 10^{-7}$</td>
<td>$2.82 \times 10^{-2}$</td>
</tr>
<tr>
<td>298</td>
<td>$8.90 \times 10^{-7}$</td>
<td>$3.66 \times 10^{-2}$</td>
</tr>
</tbody>
</table>
A linear relationship between the logarithm of the rate constant and the reciprocal of the absolute temperature was found for both the non-catalyzed and catalyzed reaction. The activation energies were found to be 47.85 kJ mol\(^{-1}\) for the catalyzed reaction and 71.77 kJ mol\(^{-1}\) for the non-catalyzed reaction. The accuracy and precision of the measurements are presented in Table II. The relative error ranged from 4.80 to 3.25 % for a Co(II) concentration in the range from 1.17 do 11.7 ng/cm\(^3\).

### Table II. Accuracy and precision of the Co(II) determination

<table>
<thead>
<tr>
<th>Taken/(ng/cm(^3))</th>
<th>Found ((\bar{x}))/(ng/cm(^3))</th>
<th>(n)</th>
<th>(S'/(ng/cm(^3)))</th>
<th>(G/%)</th>
<th>((x - \mu) \cdot 100/\mu)%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1.17</td>
<td>1.12</td>
<td>5</td>
<td>0.044</td>
<td>4.80</td>
<td>- 4.30</td>
</tr>
<tr>
<td>4.12</td>
<td>4.07</td>
<td>5</td>
<td>0.103</td>
<td>3.12</td>
<td>-1.80</td>
</tr>
<tr>
<td>11.70</td>
<td>11.88</td>
<td>5</td>
<td>0.312</td>
<td>3.25</td>
<td>1.50</td>
</tr>
</tbody>
</table>

\(\bar{x}\) – Mean value; \(\mu\) – true value; \(n\) – number of determinations; \(S'\) – standard deviation; \(G\) – relative error (=100\(\frac{t}{s/\sqrt{n}}\), where \(n = 5\) and \(t\) is Student’s for 95 % confidence)

The minimum concentration of Co(II), which could be determined by this method may be calculated by the method given by Perez–Bendito and Silva.\(^{18}\) The found detection limit is 0.20 ng/cm\(^3\).

To assess the selectivity of the method, the influence of several foreign ions on the catalytic reaction rate was examined, at a constant Co(II) concentration of 11.70 ng/cm\(^3\).

### Table III. Tolerance levels of interference in the kinetic determination of 11.70 ng/cm\(^3\) Co(II) using the optimum conditions

<table>
<thead>
<tr>
<th>Tolerance level (c(\text{Ion})/c(\text{Co(II)}))</th>
<th>Ion added</th>
</tr>
</thead>
<tbody>
<tr>
<td>(10^2)</td>
<td>Na(^+); K(^+); NH(_4)(^+); Ca(^{2+}); Ba(^{2+}); Al(^{3+}); CH(_3)COO(^-); NO(_3)(^-); VO(_2)(^+); SO(_4)(^{2-}); HPO(_4)(^{2-}); C(_2)O(_4)(^{2-}); F(^-); Cl(^-); MoO(_4)(^{2-}); Cr(_2)O(_7)(^{2-}); Zn(^{2+})</td>
</tr>
<tr>
<td>10</td>
<td>Mg(^{2+}); Li(^+); Mn(^{2+})</td>
</tr>
<tr>
<td>1</td>
<td>Fe(^{3+}); Fe(^{2+})</td>
</tr>
<tr>
<td>0.1</td>
<td>Cu(^{2+})</td>
</tr>
<tr>
<td>Interfere</td>
<td>Ni(^{2+})</td>
</tr>
</tbody>
</table>

The selectivity was determined by the 2-s criterion\(^{19}\) and the results are given in Table III, from which it may be seen that Cu(II) in a 1:1 ratio to Co(II) interfere with the reaction, Ni(II) ions interfere with this reaction at all concentrations. The other ions investigated have practically no influence on the determination of Co(II) by this method. Hence, it can be concluded that the selectivity of the method is good.

**DETERMINATION OF Co(II) IN PHARMACEUTICAL SAMPLE**

The method was directly applied for the determination of Co(II) in a pharmaceutical preparation Oligovit (Galenika a.d. Beograd). The results obtained by in-
interpolation of calibration graph are given in Table IV. The results obtained by the proposed method were compared with those obtained by atomic absorption spectrophotometry (AAS). These results show that the proposed method is applicable for the determination of Co(II) in pharmaceutical and other samples.

TABLE IV. Determination of Co(II) in a pharmaceutical sample

<table>
<thead>
<tr>
<th>Method</th>
<th>Taken Co(II) ng/cm³</th>
<th>Found Co(II) ng/cm³</th>
<th>S_e/(ng/cm³)</th>
<th>n</th>
<th>G/%</th>
<th>(x – μ) · 100/μ</th>
<th>AAS</th>
</tr>
</thead>
<tbody>
<tr>
<td>Calibration curve</td>
<td>2.94</td>
<td>2.88</td>
<td>5</td>
<td>0.13</td>
<td>12.78</td>
<td>–2.00</td>
<td>2.94</td>
</tr>
</tbody>
</table>

For the determination of cobalt in pharmaceutical preparations, the tablet was stirred for 2–3 min in deionized water, the mixture was made up to 50 cm³ with water and then filtered through a Whatman No1 filter paper. The filtered solution was diluted with water, if necessary, depending on the cobalt content.

The integral variant of the tangent method was also applied for the determination of Co(II) in the pharmaceutical preparation specified above. The results are shown in Table IV. The reproducibility was very good. The results obtained by the proposed method also agreed well with the values claimed on the labels.

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