COMPARATIVE ASSESSMENT OF THE ANALYTICAL PARAMETERS IN ASCORBIC ACID AND SULPHITE ASSAY AT A SPECTROGRAPHIC CARBON WORKING ELECTRODE

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Abstract

The aim of this study is the comparative investigation of spectrographic carbon electrode’s viability as working electrode, in ascorbic acid and sulphite assessment. Cyclic voltammetry involves a linear sweeping of the potential, the analytical signal being represented by the anodic oxidation /cathodic reduction peak of the analyte. For both analytes, the electro-oxidation resulted in an anodic peak, correlable with ascorbic acid / sulphite concentration. The analytical range of linear response corresponded to 0.07 - 10 mM for ascorbic acid and to 15.5 mg/L - 4 g/L for sulphite. The relative standard deviation RSD (%) was 2.71 % for ascorbic acid and 2.88 % for sulphite. The sensitivities, given by the slopes of the calibration graphs were 88.88 μA/m mole/L for ascorbic acid and 477.37 μA/g/L for sulphite.

Keywords: electro-oxidation, cyclic voltammetry, carbon electrode, ascorbic acid, sulphite.

INTRODUCTION

Microbial contamination and lipid oxidation are major problems threatening public health, hence there is an increasing interest in characterizing and analysing products that act against microbial spoilage and oxidative decay (Ghaderi-Ghahfarokhi et al., 2016; Nichita et al. 2016; Ghiduruş et al., 2017).

Sulphite represents a preservation agent effectively acting as microbial inhibitor, mainly in acid or acidified food (Splittoesser & Mattick, 1981; McFeeters, 1998). Sulfite can act directly through enzyme (polyphenol oxidase) inhibition, or by reducing o-quinone derivatives to 1,2-dihydroxybenzene derivatives that are more stable, hence inhibition of browning is achieved at early stage (Lesschaeve & Noble, 2005; Bushnell et al., 2003).

Ascorbic acid is a hydrosoluble antioxidant vitamin, that can scavenge singlet oxygen, act as a chelating agent, and trap hydroxyl, alkoxyl, superoxide or tocopheroxyl radicals (Du et al., 2012). Nevertheless, prooxidant activity has been reported in the presence of heavy metal cations and in the absence of other antioxidant compounds, such as SO2 (Bradshaw et al., 2011).

Chromatographic techniques, such as HPLC with UV detection were applied in the assay of both ascorbic acid and sulphite (Rodrı́guez-Comesaña et al., 2002; McFeeters & Barish, 2003). Spectrophotometry was also applied to the assay of these key compounds for food and beverage industries (Brychkova et al., 2012; Kapur et al., 2012).

Electrochemical techniques have gained increasing interest in food analysis and clinical domain. They are characterized by sensitivity and selectivity, rapidity, as well as by simplicity of the applied procedure, requiring no laborious sample pre-treatment and involving relatively low cost, when compared to conventional instrumental methods. Therefore they were applied to quantitative assessment of food additives and ingredients (Bard & Faulkner, 1980; Pisoschi et al., 2006).

Voltammetry involves recording of the current intensity, at a controlled potential variation. In the case of irreversible redox couples, such as those confirmed for ascorbic acid and sulphite, the intensity of the cyclic voltammetric anodic oxidation peak can be directly correlated to the analyte concentration. Sulphite is irreversibly oxidized to sulphate (Lu et al., 1999; Skavas & Hemmingsen, 2007; Pisoschi, 2017), whereas...
oxidation of ascorbic acid to dehydroascorbic acid is followed by an irreversible solvation reaction at pH<4, yielding 2,3-diketogulonic acid, electroinactive (Erdurak-Kiliç et al., 2006; Pisoschi et al., 2014).

From bare to modified electrodes based on advanced materials, good analytical performances with respect to accuracy and sensitivity in ascorbic acid and sulphite assay, were obtained (Erdurak-Kiliç et al., 2006; Pisoschi, 2014; Pisoschi et al, 2014).

A novel poly(2-(N-morpholine) ethane sulfonic acid)/reduced graphene oxide modified glassy carbon electrode was developed, by electropolymerization. Cyclic and differential pulse voltammetry were applied, to investigate the electrochemical behaviors and to simultaneously detect ascorbic acid, dopamine and uric acid. The electrode exhibited enhanced electrocatalytic activity towards the oxidation of three analytes, with detection limits of 0.43 μM, 0.0062 μM and 0.056 μM for ascorbic acid, dopamine and uric acid, respectively (Zhang et al., 2018).

Cyclic voltammetry at glassy carbon electrode was performed scanning the potential from -1.0 to 1.0 V vs. Ag/AgCl, at a 100 mV s⁻¹ scan rate, developing a calibration curve from 10⁻⁴ mol L⁻¹ to 10⁻¹ mol L⁻¹ sodium sulphite in basic solution, NaOH 0.02 M (Arce et al. 2016).

The present study aims at the comparative investigation of the analytical parameters, in ascorbic acid and sulphite determination at the spectrographic carbon working electrode.

**MATERIALS AND METHODS**

A KSP potentiostat-galvanostat, laboratory made by Professor Slawomir Kalinowski, University Warmia and Mazury (Olsztyn), as well as the respective software Cyclic Voltammetry, were used for recording the cyclic voltammograms.

A working electrode made of spectrographic carbon (Topolcany, Slovakia) was used. The counter electrode was a Pt strip Radelkis OP-0612P electrode. As reference, a saturated calomel electrode (SCE), Radelkis (Hungary) was used.

The stock solution of ascorbic acid 100 mM was prepared daily by dissolving the corresponding ascorbic acid (Merck, ACS ISO) amount in a 0.10 M KCl electrolyte solution.

The stock solution of sodium sulphite (5 g/L) was prepared daily by dissolving Na₂SO₃ (Merck, ACS ISO) in a 0.10 M KCl (Chimopar, Bucharest, Romania) electrolyte solution. Standard solutions of ascorbic acid, with concentrations comprised between 0.05 mM and 10 mM were obtained, by dilution of the stock solution with 0.10 M KCl. Standard solutions of sulphite (as Na₂SO₃), with concentrations comprised between 5 mg/L and 5 g/L were obtained by dilution of the stock solution with 0.10 M KCl.

The volume of the analysed sample was 50 mL and all measurements were performed at 23°C. Before each determination, the working electrode was cleaned mechanically and electrochemically by applying a -1.5 V potential pulse for 3 seconds. For the cyclic voltammetric measurements, the potential was scanned within the range -100 - 1500 mV, with a 50 mV/s scan rate. For the investigation of the influence of the scan rate, the potential sweep rate varied between 25 and 250 mV/s.

**RESULTS AND DISCUSSIONS**

Cyclic voltammograms were recorded using the spectrographic carbon electrode, for both target analytes. Ascorbic acid concentrations ranged between 0.01 and 10 mM, and sulphite concentrations were comprised between 5 mg/L and 5 g /L. Illustrative cyclic voltammograms exhibiting only the anodic peak are presented in Figures 1 and 2.

![Figure 1: Cyclic voltammogram recorded with the spectrographic carbon working electrode, for 5 mM ascorbic acid concentration, prepared in 0.1 M KCl electrolyte solution](image-url)
ascorbic acid, as well as for sodium sulphite, both analytes being dissolved in 0.1 M KCl electrolyte solution.

The current intensity recorded for the peak height was plotted against concentration for ascorbic acid, as well as for sodium sulphite, both analytes being dissolved in 0.1 M KCl electrolyte solution.

The developed calibration curves (Figure 3 and 4) showed a linear range of analytical response corresponding to 0.07 - 10 mM for ascorbic acid and to 0.0155 g/L - 4 g/L for sulphite.

The equation of the calibration graph corresponded to \( y = 88.882 \times + 71.766 \); \( R^2=0.9902 \) (ascorbic acid) and to \( y = 477.37 \times + 257.41 \); \( R^2 = 0.9747 \) (sulphite).

The values of the relative standard deviation RSD were 2.71 % for ascorbic acid and 2.88 % for sulphite. The RSD values were calculated as:

\[
RSD(\%) = 100 \times \frac{\text{standard deviation}}{\text{the mean of determinations}}
\]

\( c = 2.5 \text{ mM, } n = 10 \text{ determinations for ascorbic acid; } c = 125 \text{ mg/L, } n = 10 \text{ determinations for sulphite.} \)

The sensitivities, given by the slopes of the calibration graphs were 88.88 \( \mu A/mM \) for ascorbic acid and 477.37 \( \mu A/g/L \) (60.10 \( \mu A/mM \)) for sulphite.

The obtained detection limits were 0.026 mM ascorbic acid and 4.85 mg/L (0.0384 mM) sulphite, calculated as LOD = 3 s/m, where s represents the square mean error calculated for the KCl electrolyte solution as blank, and m represents the slope of the calibration graph.

The current intensity corresponding to the anodic peak height depended linearly on the square root of the potential scan rate (Figure 5), observing Randles-Sevcik dependence and confirming that the process is controlled by analyte diffusion.

In Table 1, the results obtained at ascorbic acid determination in citrus juices freshly obtained by fruit squeezing, are presented.
Table 1: Results obtained at ascorbic acid (AA) assay by cyclic voltammetry at spectrographic carbon electrode. The results represent the average of 5 determinations

<table>
<thead>
<tr>
<th>Analysed product</th>
<th>AA concentration (mg/100 mL)</th>
<th>AA concentration (mM)</th>
<th>AA added amount (mg/100 mL)</th>
<th>Degree of recovery %</th>
</tr>
</thead>
<tbody>
<tr>
<td>Orange juice I</td>
<td>35.47 ± 1.19</td>
<td>2.01 ± 0.067</td>
<td>20</td>
<td>98.12</td>
</tr>
<tr>
<td>Orange juice II</td>
<td>46.98 ± 1.16</td>
<td>2.66 ± 0.066</td>
<td>25</td>
<td>99.07</td>
</tr>
<tr>
<td>Orange juice III</td>
<td>53.57 ± 1.80</td>
<td>3.04 ± 0.102</td>
<td>25</td>
<td>102.03</td>
</tr>
<tr>
<td>Lemon juice I</td>
<td>79.08 ± 2.43</td>
<td>4.49 ± 0.138</td>
<td>40</td>
<td>102.71</td>
</tr>
<tr>
<td>Lemon juice II</td>
<td>56.30 ± 1.67</td>
<td>3.2 ± 0.095</td>
<td>25</td>
<td>101.42</td>
</tr>
</tbody>
</table>

CONCLUSIONS

The spectrographic carbon working electrode proved its viability in both ascorbic acid and sulphite assay. Both analytes underwent irreversible electro-oxidation, the voltammograms exhibiting only the anodic peak. Sulphite determination was characterized by a broader linear range (15.5 mg/L - 4 g/L) than ascorbic acid assessment (0.07 - 10 mM). Nevertheless, moderately improved precision given by the relative standard deviation (2.71% versus 2.88%), better correlation coefficient (0.9902 vs 0.9747) and lower detection limit (0.026 mM vs 0.0384 mM) are obtained at ascorbic acid determination. The accuracy of the assay in real samples was proved by good values of the degrees of recovery of added analyte amounts.

REFERENCES


Splittoesser D.F., Mattick L.R., 1981. The storage life of refrigerated grape juice containing various levels of sulfur dioxide. American Journal of Enology and Viticulture, 32: 171-173