Development of indirect UV spectrophotometric method for the determination of vitamin C content in *Prosopis juliflora* and *Balanites aegyptica* using standard ceric (IV) sulphate

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Abstract:
In the present work three ultraviolet spectrophotometric methods of analysis were developed depending principally on the measurement of absorbance of ceric (IV) ions in 5M H$_2$SO$_4$ at 323nm. The first, for the direct determination of ceric(IV) ions, whereas the second, for the indirect determination of vitamin C but also through the measurement of the absorbance of the unreduced standard ceric(IV) ions after the addition of standard vitamin C. The third, applied the second method to determine vitamin C content in the extract of fruits (385ppm) and leaves (485ppm) of *Balanites aegyptica* (laloub) and of fruits (216ppm) and leaves (315ppm) of *Prosopis juliflora* (mesquite).

Key words: *Balanites aegyptica*, *Prosopis juliflora*, laloub, mesquite, ascorbic acid, ceric (IV) sulphate, spectrophotometric method.

1. INTRODUCTION

Various analytical methods have been employed for the determination of ascorbic acid in different matrices including
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Titrmetricity, voltammetry [1]-[6], liquid chromatography [1]-[9], complexometry [3], spectrophotometry, amperometry and enzymatic methods [3]-[10]-[11].

Titrimetric methods have been commonly used in the determination of vitamin C in fruit samples because they are simple; however, difficulties were encountered with commonly used titrants and interferences also occurred with colored samples [1].

Prosopis juliflora (mesquite) was introduced into Sudan since 1917 and planted in Khartoum. Its ability to tolerate drought and fix sand dunes provided the impetus for the introduction of the tree into various parts of Sudan (central, northern and eastern Sudan) with emphasis on dry areas [12]. Balanites aegyptiaca (laloub) is also widely distributed in Sudan such as Kordofan and Darfur [13].

Fruits and leaves of Balanites aegyptiaca and Prosopis juliflora, are expected to contain micro- and macro- nutrients, minerals and vitamins including vitamin C.

No investigations in the methods of extraction and determination of vitamin C in fruits and leaves of Balanites aegyptiaca and Prosopis juliflora in Sudan have been reported.

In the present work various methods of extraction of vitamin C, from fruits and leaves of Balanites aegyptiaca and Prosopis juliflora, and the determination of its content by indirect UV- spectrophotometry using ceric (VI) sulphate will be investigated.

2. Materials and methods

2.1 Chemicals

All chemicals used were of analytical grade.

- L (+) Ascorbic acid powder (ANKARA .TORKIYE).
- Ceric sulphate, Ce(SO₄)₂ 2H₂O, 99.9% pure, BDH Poole England.
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- Sulphuric acid (INDIA).
- Metaphosphoric acid (Sigma-Aldrich (Johannesburg, South Africa)).
- Acetic acid (Fluka) (chemika).

2.2 Equipment

- UV/VIS spectro star Nano (BMG LABTECH) with 1-cm matched quartz cells.

2.3 Samples

Both leaves of *Balanites aegyptica* (BL) and *Prosopis juliflora* (PL) and fruits of the latter (PF) were collected from the trees grown in the different parts of Khartoum State (Sudan). However, fruits of the former (BF) were purchased from Khartoum State market.

2.4 Preparation of solutions

**Ceric (IV) sulphate solution (1000 ppm)**

0.1g of ceric (IV) sulphate (Ce (SO₄)₂ 2H₂O) was weighed accurately, dissolved in 5M H₂SO₄ and diluted to 100 ml with distilled water.

**Ascorbic acid (100 ppm)**

0.01g ascorbic acid (A.R.) was weighed accurately, dissolved and diluted to 100ml with distilled water.

**Blank**

0.5ml 5M H₂SO₄ was diluted to 50 ml with distilled water.

2.5 Extraction methods of vitamin C

1 g of each of the fruits and leaves was weighed and crushed, to which, 25 ml of each solvents (distilled water, 4.5% metaphosphoric acid or acetic acid) was added and after 30 minutes the contents were squeezed and filtered.
2.6 Optimization of the experimental conditions
The effect of various parameters comprising wavelength of maximum absorption, temperature and temperature was investigated. Absorbance of ceric (IV) sulphate was measured at five wavelengths (300, 323, 325, 332 and 340nm) and the maximum absorbance was obtained at 323nm. The absorbance of ceric(IV) sulphate was also measured in 4M, 5M and 6M H₂SO₄. The maximum absorbance was obtained in 5M H₂SO₄.

3. RESULTS AND DISCUSSION

3.1 UV spectrophotometric method for the determination of ceric(IV) sulphate
Different concentrations of ceric (IV) sulphate in 5M H₂SO₄ were prepared and their absorbance at 323nm was recorded in Table 1 and drawn in Figure 1.

Table 1 Absorbance of ceric (IV) sulphate standard in 5M H₂SO₄ at 323nm

<table>
<thead>
<tr>
<th>Ceric (IV)sulphate concentration ppm</th>
<th>20</th>
<th>40</th>
<th>60</th>
<th>80</th>
<th>100</th>
<th>120</th>
<th>140</th>
<th>150</th>
</tr>
</thead>
<tbody>
<tr>
<td>Absorbance</td>
<td>0.182</td>
<td>0.380</td>
<td>0.592</td>
<td>0.782</td>
<td>0.999</td>
<td>1.231</td>
<td>1.451</td>
<td>1.647</td>
</tr>
</tbody>
</table>

Figure 1 Beer’s law plot of ceric (IV) sulphate standard in 5M H₂SO₄ at 323nm
3.2 Indirect UV spectrophotometric determination of ascorbic acid in *Balanites egyptica* and *Prosopis juliflora* using ceric (IV) sulphate

1 ml of each vitamin C filtrate (using distilled water, 4.5% metaphosphoric acid or acetic acid) was pipetted into 50 ml volumetric flask, and 5 ml ceric (IV) sulphate (150 ppm) was added. This was then completed to the mark with distilled water. Absorbance of 150 ppm ceric (IV) sulphate, in 5M H$_2$SO$_4$ which was not reduced after the addition of varying volumes of 100 ppm ascorbic acid was recorded in Table 2 and Figure 2.

Table 2 Absorbance of ceric(IV) sulphate after addition of ascorbic acid

<table>
<thead>
<tr>
<th>Ascorbic acid volumes (ml)</th>
<th>Absorbance at 323nm</th>
</tr>
</thead>
<tbody>
<tr>
<td>0</td>
<td>0.781</td>
</tr>
<tr>
<td>0.2</td>
<td>0.725</td>
</tr>
<tr>
<td>0.4</td>
<td>0.653</td>
</tr>
<tr>
<td>0.6</td>
<td>0.590</td>
</tr>
<tr>
<td>0.8</td>
<td>0.536</td>
</tr>
<tr>
<td>1</td>
<td>0.464</td>
</tr>
<tr>
<td>1.2</td>
<td>0.396</td>
</tr>
<tr>
<td>1.4</td>
<td>0.334</td>
</tr>
<tr>
<td>1.6</td>
<td>0.275</td>
</tr>
<tr>
<td>1.8</td>
<td>0.236</td>
</tr>
<tr>
<td>2</td>
<td>0.157</td>
</tr>
<tr>
<td>2.2</td>
<td>0.103</td>
</tr>
<tr>
<td>2.4</td>
<td>0.051</td>
</tr>
<tr>
<td>2.6</td>
<td>0.028</td>
</tr>
</tbody>
</table>

Figure 2 Beer’s law plot of ascorbic acid solutions at 323nm
3.3 Efficiency of the extracting solutions

Distilled water, metaphosphoric acid and acetic acid were used to extract vitamin C from fruits or leaves of *Balanites aegyptica* and *Prosopis juliflora*.

Vitamin C concentration was determined in these extracts and the results obtained are shown in Table 3.

Table 3 Ascorbic acid contents in fruits and leaves of *Balanites aegyptica* and *Prosopis juliflora*.

<table>
<thead>
<tr>
<th>Common name</th>
<th>Botanical name</th>
<th>Ascorbic acid conc extracted with distilled. water ppm</th>
<th>Ascorbic acid conc extracted with M. phosphoric acid ppm</th>
<th>Ascorbic acid conc extracted with acetic acid ppm</th>
</tr>
</thead>
<tbody>
<tr>
<td>Laloub</td>
<td><em>Balanites aegyptica</em> F</td>
<td>326</td>
<td>380</td>
<td>361</td>
</tr>
<tr>
<td>Laloub</td>
<td><em>Balanites aegyptica</em> L</td>
<td>423</td>
<td>465</td>
<td>441</td>
</tr>
<tr>
<td>Mesquite</td>
<td><em>Prosopis juliflora</em> F</td>
<td>179</td>
<td>216</td>
<td>198</td>
</tr>
<tr>
<td>Mesquite</td>
<td><em>Prosopis juliflora</em> L</td>
<td>283</td>
<td>315</td>
<td>306</td>
</tr>
</tbody>
</table>

Metaphosphoric acid was the most efficient extracting solution for both leaves (465ppm, 315ppm) and fruits (380ppm, 216ppm) of *Balanites aegyptica* and *Prosopis juliflora*, respectively. Distilled water was the least, for leaves (423ppm, 283ppm) and fruits (326ppm, 179ppm), respectively. In between, acetic acid showed middle efficiency (441ppm, 306ppm) for leaves and (361ppm, 198ppm) for fruits, of *Balanites aegyptica* and *Prosopis juliflora*, respectively. The success attained in development of an indirect UV spectrophotometric determination of vitamin C content in laloub and mesquite tempts to investigate the application of the method for the determination of other constituents in other dry food materials using even indirect other instrumental techniques.
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REFERENCES


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