Supplementary Information

Method Development and Total Uncertainty Estimation for Boron, Sulfur and Phosphorus Determination in Mineral Fertilizer Using ICP OES

Tiago C. de Oliveira, Roger T. Rampazzo, Marco F. Ferrão and Dirce Pozebon

Laboratório Nacional Agropecuário do Rio Grande do Sul, Ministério da Agricultura Pecuária e Abastecimento, Estrada da Ponta Grossa 3036, 91780-580 Porto Alegre-RS, Brazil

Instituto de Química, Universidade Federal do Rio Grande do Sul, 91501-970 Porto Alegre-RS, Brazil

The following procedure was transcribed from the Official Methods of the Brazilian Ministry of Agriculture, Livestock and Supply (Ministério da Agricultura, Pecuária e Abastecimento (MAPA)).

Preparation of neutral ammonium citrate solution (NAC)

Dissolve 370 g of monohydrate citric acid (C₆H₈O₇.H₂O) in 1500 mL of water and add 345 mL of ammonium hydroxide (NH₄OH), containing 28 to 29% of NH₃. If the NH₃ concentration is lower than 28%, add more NH₄OH to compensate for the lower NH₃ concentration. Decrease the volume of water (the same volume of the additional aliquot of NH₄OH) in which citric acid is dissolved. Allow the solution to cool and then measure its pH. If necessary, adjust the pH to 7.0 ± 0.05 with NH₄OH solution (NH₄OH 8-fold diluted in water) or 10% (m/v) citric acid solution. Store the NAC solution in closed flask and check the pH weekly or when necessary.

Extraction of soluble P using NAC

(i) Weigh 1 g of sample with 0.1 mg of precision and transfer to filter paper with medium porosity, adapted in a funnel placed on a volumetric 500 mL flask.
(ii) Wash the sample with 180 mL of water by adding small portions.
(iii) Transfer the filter paper with the residue to a 250-300 mL Erlenmeyer flask and rinse the funnel with water, while transferring to the 500 mL volumetric flask.
(iv) Add 100 mL of NAC previously heated at 65 °C to the Erlenmeyer flask.
(v) Cap the flask and shake the mixture vigorously for a few minutes. Eventually, remove the cap for pressure release.
(vi) Place the flask tightly closed in oven or water bath with stirrer and stir for 1 hour, keeping the temperature at 65° ± 5 °C;
(vii) After 1 h, remove the flask from the shaking system and leave the mixture to cool to room temperature. Subsequently, transfer the mixture to the flask containing the water-soluble phosphorus. Complete the volume of the flask and mix the solution.
(viii) Leave the mixture standing until a clear supernatant is observed. Then, filter the supernatant (medium porosity filter) or centrifuge it.

*e-mail: dircepoz@iq.ufrgs.br
Table S1. Maximum permissible errors for volumetric flasks, pipettes and volumetric micropipettes

<table>
<thead>
<tr>
<th>Calibrated item</th>
<th>Maximum tolerated error&lt;sup&gt;a&lt;/sup&gt;</th>
</tr>
</thead>
<tbody>
<tr>
<td>Volumetric flask (50 mL)</td>
<td>0.12 mL</td>
</tr>
<tr>
<td>Volumetric flask (100 mL)</td>
<td>0.20 mL</td>
</tr>
<tr>
<td>Volumetric flask (200 and 250 mL)</td>
<td>0.30 mL</td>
</tr>
<tr>
<td>Volumetric flask (500 mL)</td>
<td>0.50 mL</td>
</tr>
<tr>
<td>Volumetric pipette (1 mL)</td>
<td>0.014 mL</td>
</tr>
<tr>
<td>Volumetric pipette (2 and 3 mL)</td>
<td>0.02 mL</td>
</tr>
<tr>
<td>Volumetric pipette (4 to 7 mL)</td>
<td>0.03 mL</td>
</tr>
<tr>
<td>Volumetric pipette (8 to 10 mL)</td>
<td>0.04 mL</td>
</tr>
<tr>
<td>Volumetric pipette (15 to 25 mL)</td>
<td>0.06 mL</td>
</tr>
<tr>
<td>Volumetric pipette (50 mL)</td>
<td>0.10 mL</td>
</tr>
<tr>
<td>Micropipette (50 µL)</td>
<td>0.50 µL</td>
</tr>
<tr>
<td>Micropipette (100 µL)</td>
<td>0.80 µL</td>
</tr>
<tr>
<td>Micropipette (200 µL)</td>
<td>1.6 µL</td>
</tr>
<tr>
<td>Micropipette (500 µL)</td>
<td>4.0 µL</td>
</tr>
<tr>
<td>Micropipette (1000 µL)</td>
<td>8.0 µL</td>
</tr>
</tbody>
</table>

<sup>a</sup>Quadratic sum of the expanded uncertainty and the measurement error.

Doehlert matrix

A Doehlert matrix with three variables can be represented as a cube-octahedron.<sup>1</sup> The exact composition of the experimental array, with the definition of values used for each experiment, may change with the cube-octahedron spin.<sup>1,2</sup> Figure S1 illustrates a possible visualization of the composition for the present study, whereas the numeric values of the coordinates are given in Table S2. The values of the coordinates were converted into real values. A dimension of the spatial model was assigned to a studied factor; the zero coordinate in the space corresponded to the average value of the experimental range for the factor. Then, the distance from the average to the maximum value of this range was determined. This distance is equivalent to the radius of the sphere containing the cube-octahedron. Thus, the average value plus the value of the radius of the sphere was the highest in the range considered. Coordinate +1 was attributed to this value, whereas the minimum value of the range was assigned to coordinate –1.
**Figure S1.** Experimental Doehlert design. (Top) Model in perpendicular view to the axis $X_3$. (Bottom) Scheme of a three dimensional view. The geometric representation of numbered points corresponds to each test number in Table S2. $C_1$ to $C_n$ are the repetitions of the central point. $X_1$, $X_2$ and $X_3$ represent the first, the second and third experimental variable, respectively.$^3$
<table>
<thead>
<tr>
<th>Assay</th>
<th>$X_1$</th>
<th>$X_2$</th>
<th>$X_3$</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>1</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>2</td>
<td>0.5</td>
<td>0.866</td>
<td>0</td>
</tr>
<tr>
<td>3</td>
<td>0.5</td>
<td>0.289</td>
<td>0.817</td>
</tr>
<tr>
<td>4</td>
<td>-1</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>5</td>
<td>-0.5</td>
<td>-0.866</td>
<td>0</td>
</tr>
<tr>
<td>6</td>
<td>-0.5</td>
<td>-0.289</td>
<td>-0.817</td>
</tr>
<tr>
<td>7</td>
<td>-0.5</td>
<td>-0.866</td>
<td>0</td>
</tr>
<tr>
<td>8</td>
<td>0.5</td>
<td>-0.289</td>
<td>-0.817</td>
</tr>
<tr>
<td>9</td>
<td>0.5</td>
<td>0.866</td>
<td>0</td>
</tr>
<tr>
<td>10</td>
<td>-0.5</td>
<td>0.577</td>
<td>-0.817</td>
</tr>
<tr>
<td>11</td>
<td>0</td>
<td>0.289</td>
<td>0.817</td>
</tr>
<tr>
<td>12</td>
<td>0</td>
<td>-0.577</td>
<td>0.817</td>
</tr>
<tr>
<td>$c_1$</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>$c_2$</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>...</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
<tr>
<td>$c_n$</td>
<td>0</td>
<td>0</td>
<td>0</td>
</tr>
</tbody>
</table>

$X_1$, $X_2$ and $X_3$ represent the first, the second and third experimental variable, respectively.
Table S3. Factors and levels evaluated in optimizing the instrumental conditions and results obtained

<table>
<thead>
<tr>
<th>Assay</th>
<th>Levels and factors</th>
<th>Intensity</th>
<th>Plasma robustness</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td>Power / kW</td>
<td>Peristaltic pump speed / rpm</td>
<td>Nebulizer gas pressure / kPa</td>
</tr>
<tr>
<td>1</td>
<td>1.45</td>
<td>15.00</td>
<td>200.00</td>
</tr>
<tr>
<td>2</td>
<td>1.30</td>
<td>24.00</td>
<td>200.00</td>
</tr>
<tr>
<td>3</td>
<td>1.30</td>
<td>18.00</td>
<td>220.00</td>
</tr>
<tr>
<td>4</td>
<td>0.75</td>
<td>15.00</td>
<td>200.00</td>
</tr>
<tr>
<td>5</td>
<td>0.90</td>
<td>6.00</td>
<td>200.00</td>
</tr>
<tr>
<td>6</td>
<td>0.90</td>
<td>12.00</td>
<td>180.00</td>
</tr>
<tr>
<td>7</td>
<td>1.30</td>
<td>6.00</td>
<td>200.00</td>
</tr>
<tr>
<td>8</td>
<td>1.30</td>
<td>12.00</td>
<td>180.00</td>
</tr>
<tr>
<td>9</td>
<td>0.90</td>
<td>24.00</td>
<td>200.00</td>
</tr>
<tr>
<td>10</td>
<td>1.10</td>
<td>21.00</td>
<td>180.00</td>
</tr>
<tr>
<td>11</td>
<td>0.90</td>
<td>18.00</td>
<td>220.00</td>
</tr>
<tr>
<td>12</td>
<td>1.10</td>
<td>9.00</td>
<td>220.00</td>
</tr>
<tr>
<td>13</td>
<td>1.10</td>
<td>15.00</td>
<td>200.00</td>
</tr>
<tr>
<td>14</td>
<td>1.10</td>
<td>15.00</td>
<td>200.00</td>
</tr>
<tr>
<td>15</td>
<td>1.10</td>
<td>15.00</td>
<td>200.00</td>
</tr>
<tr>
<td>16</td>
<td>1.10</td>
<td>15.00</td>
<td>200.00</td>
</tr>
<tr>
<td>17</td>
<td>1.10</td>
<td>15.00</td>
<td>200.00</td>
</tr>
<tr>
<td>18</td>
<td>1.10</td>
<td>15.00</td>
<td>200.00</td>
</tr>
</tbody>
</table>

\(R^2\): coefficient of determination.
Table S4. Uncertainty contribution factors and sensitivity coefficients

<table>
<thead>
<tr>
<th>Uncertainty component</th>
<th>Sensitivity coefficient</th>
</tr>
</thead>
<tbody>
<tr>
<td>L₁ (curve calibration regression)</td>
<td>$\frac{FD}{10000 \times m}$</td>
</tr>
<tr>
<td>Analyte concentration (S)</td>
<td>L₂ (standard total error) $\frac{FD}{PC}$</td>
</tr>
<tr>
<td></td>
<td>L₃ (standard dilutions) $- \frac{CP}{PC^2} \times FD$</td>
</tr>
<tr>
<td>Dilution factor (DF)</td>
<td>DF (maximum tolerated error (including volumetric expansion of glassware)) $\frac{L}{10000 \times m}$</td>
</tr>
<tr>
<td>Sample mass (m)</td>
<td>m₁ (balance total error)</td>
</tr>
<tr>
<td></td>
<td>m₂ (balance resolution) $\frac{L \times FD}{10000m^2}$</td>
</tr>
<tr>
<td></td>
<td>m₃ (eccentricity)</td>
</tr>
<tr>
<td>R&amp;R (intralaboratory reproducibility)</td>
<td>1</td>
</tr>
<tr>
<td>REC (average recovery)</td>
<td>1</td>
</tr>
</tbody>
</table>

PC: dilution of the solution with the highest concentration in the calibration curve; CP: concentration of the analyte in calibration solution.

Table S5. Contributions of each component to the uncertainty in the determination of B, S, and P in mineral fertilizer using ICP OES and analytes recovery

<table>
<thead>
<tr>
<th>Uncertainty components</th>
<th>B</th>
<th>S</th>
<th>P</th>
<th>Degrees of freedom</th>
</tr>
</thead>
<tbody>
<tr>
<td>Sample Concentration / % (m/m)</td>
<td>0.232</td>
<td>5.87</td>
<td>7.11</td>
<td>–</td>
</tr>
<tr>
<td>Analyte concentration (S)</td>
<td>L₁(curve regression) 0.0061</td>
<td>0.2667</td>
<td>0.6077</td>
<td>16</td>
</tr>
<tr>
<td></td>
<td>L₂ (standard total error) 0.00111</td>
<td>0.2494</td>
<td>0.1386</td>
<td>$\infty$</td>
</tr>
<tr>
<td></td>
<td>L₃ (standard dilutions) 31.5352</td>
<td>0.2090</td>
<td>0.2014</td>
<td>$\infty$</td>
</tr>
<tr>
<td>Dilution factor (DF)</td>
<td>DF (maximum tolerated error) 204.3333</td>
<td>3.1749</td>
<td>17.3181</td>
<td>$\infty$</td>
</tr>
<tr>
<td></td>
<td>m₁ (balance total error) 0.0003</td>
<td>0.0005</td>
<td>0.0003</td>
<td>$\infty$</td>
</tr>
<tr>
<td>Sample mass (m)</td>
<td>m₂ (balance resolution) 0.00005</td>
<td>0.017</td>
<td>0.0184</td>
<td>25</td>
</tr>
<tr>
<td></td>
<td>m₃ (eccentricity) 0.0002</td>
<td>0.0002</td>
<td>0.0184</td>
<td>29</td>
</tr>
<tr>
<td>Intralaboratory reproducibility / %</td>
<td>0.0279</td>
<td>0.017</td>
<td>0.0184</td>
<td>25</td>
</tr>
<tr>
<td>Accuracy / %</td>
<td>102.35</td>
<td>100</td>
<td>103.58</td>
<td>29</td>
</tr>
</tbody>
</table>
Table S6. Data obtained in the experiments of the laboratory reproducibility evaluation

<table>
<thead>
<tr>
<th>Analyte</th>
<th>Sample</th>
<th>Repetition</th>
<th>Measurement 1</th>
<th>Measurement 2</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>A</td>
<td>0.0724</td>
<td>0.0703</td>
</tr>
<tr>
<td></td>
<td>B</td>
<td>0.0736</td>
<td>0.0690</td>
<td></td>
</tr>
<tr>
<td></td>
<td>C</td>
<td>0.0711</td>
<td>0.0689</td>
<td></td>
</tr>
<tr>
<td></td>
<td>A</td>
<td>10.77</td>
<td>10.64</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>B</td>
<td>10.51</td>
<td>10.72</td>
</tr>
<tr>
<td></td>
<td>C</td>
<td>10.64</td>
<td>10.53</td>
<td></td>
</tr>
<tr>
<td></td>
<td>A</td>
<td>0.0415</td>
<td>0.0424</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>B</td>
<td>0.0418</td>
<td>0.0406</td>
</tr>
<tr>
<td></td>
<td>C</td>
<td>0.0407</td>
<td>0.0436</td>
<td></td>
</tr>
<tr>
<td></td>
<td>A</td>
<td>1.027</td>
<td>1.131</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>B</td>
<td>1.030</td>
<td>1.051</td>
</tr>
<tr>
<td></td>
<td>C</td>
<td>1.019</td>
<td>1.038</td>
<td></td>
</tr>
<tr>
<td></td>
<td>A</td>
<td>0.2319</td>
<td>0.2445</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>B</td>
<td>0.2357</td>
<td>0.2448</td>
</tr>
<tr>
<td></td>
<td>C</td>
<td>0.2427</td>
<td>0.2361</td>
<td></td>
</tr>
<tr>
<td></td>
<td>A</td>
<td>13.70</td>
<td>13.31</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>B</td>
<td>13.66</td>
<td>13.26</td>
</tr>
<tr>
<td></td>
<td>C</td>
<td>14.13</td>
<td>13.48</td>
<td></td>
</tr>
<tr>
<td></td>
<td>A</td>
<td>10.86</td>
<td>11.08</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>B</td>
<td>10.84</td>
<td>11.05</td>
</tr>
<tr>
<td></td>
<td>C</td>
<td>10.99</td>
<td>10.98</td>
<td></td>
</tr>
<tr>
<td></td>
<td>A</td>
<td>2.057</td>
<td>2.127</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>B</td>
<td>2.128</td>
<td>2.165</td>
</tr>
<tr>
<td></td>
<td>C</td>
<td>2.083</td>
<td>2.125</td>
<td></td>
</tr>
<tr>
<td></td>
<td>A</td>
<td>6.85</td>
<td>7.02</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>B</td>
<td>7.08</td>
<td>6.94</td>
</tr>
<tr>
<td></td>
<td>C</td>
<td>7.13</td>
<td>7.06</td>
<td></td>
</tr>
<tr>
<td></td>
<td>A</td>
<td>5.747</td>
<td>5.872</td>
<td></td>
</tr>
<tr>
<td></td>
<td></td>
<td>B</td>
<td>5.925</td>
<td>5.891</td>
</tr>
<tr>
<td></td>
<td>C</td>
<td>5.894</td>
<td>6.003</td>
<td></td>
</tr>
</tbody>
</table>
Table S6. Data obtained in the experiments of the laboratory reproducibility evaluation (cont.)

<table>
<thead>
<tr>
<th>Analyte</th>
<th>Sample</th>
<th>Repetition</th>
<th>Measurement 1</th>
<th>Measurement 2</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>A</td>
<td>10.69</td>
<td>11.02</td>
</tr>
<tr>
<td>Phosphorus (213.547 nm)</td>
<td>0162F/15</td>
<td>B</td>
<td>11.03</td>
<td>11.31</td>
</tr>
<tr>
<td></td>
<td></td>
<td>C</td>
<td>10.96</td>
<td>11.26</td>
</tr>
<tr>
<td></td>
<td></td>
<td>A</td>
<td>13.40</td>
<td>13.58</td>
</tr>
<tr>
<td></td>
<td></td>
<td>B</td>
<td>13.70</td>
<td>13.71</td>
</tr>
<tr>
<td></td>
<td></td>
<td>C</td>
<td>13.45</td>
<td>13.79</td>
</tr>
<tr>
<td></td>
<td></td>
<td>A</td>
<td>5.63</td>
<td>5.56</td>
</tr>
<tr>
<td></td>
<td>1197F/14</td>
<td>B</td>
<td>5.53</td>
<td>5.60</td>
</tr>
<tr>
<td></td>
<td></td>
<td>C</td>
<td>5.35</td>
<td>5.59</td>
</tr>
<tr>
<td></td>
<td></td>
<td>A</td>
<td>7.11</td>
<td>7.28</td>
</tr>
<tr>
<td></td>
<td>0186F/15</td>
<td>B</td>
<td>7.26</td>
<td>7.52</td>
</tr>
<tr>
<td></td>
<td></td>
<td>C</td>
<td>7.37</td>
<td>7.54</td>
</tr>
<tr>
<td></td>
<td></td>
<td>A</td>
<td>6.49</td>
<td>6.68</td>
</tr>
<tr>
<td></td>
<td>0213F/15</td>
<td>B</td>
<td>6.61</td>
<td>6.52</td>
</tr>
<tr>
<td></td>
<td></td>
<td>C</td>
<td>6.72</td>
<td>6.57</td>
</tr>
</tbody>
</table>

The normalized error informed in Table 9 was calculated according to the following equation:

\[
E_n = \left( \frac{x_{lab} - X_{reference}}{\sqrt{U_{lab}^2 + U_{reference}^2}} \right)
\]  

(1)

U = expanded uncertainty  
E_n = normalized error  
E_n < 1 to pass

References