

Estimates of Precision in a Standard Additions Analysis

Graham R. Bruce*

Department of Chemistry, Okanagan University College, Kelowna, BC, Canada, V1V 1V7; *grbruce@okanagan.bc.ca

Paramjit S. Gill

Department of Mathematics and Statistics, Okanagan University College, Kelowna, BC, Canada, V1V 1V7; pgill@okanagan.bc.ca.

In an instrumental analysis course the students carried out a spectrophotometric analysis using the method of standard additions to overcome matrix interferences. As well as determining the concentration of a supplied unknown, the students were required to estimate the precision of their analysis using the 95% confidence interval, $c_x \pm t s_c$. Here, c_x is the experimentally determined analyte concentration, t a value obtained from a Student t -table, and s_c an estimate of the standard error in c_x .

It became apparent that the students, having used different textbook sources, were applying two methods to calculate s_c and these yielded different values. The purpose of this article is to identify the source of this disagreement and to rectify the problem.

Preliminaries

An experimental procedure for a standard additions analysis (I) is as follows. Equal volumes, V_x , of an analyte solution of unknown concentration, c_x , are added to a series of volumetric flasks of volume V_t . A standard analyte solution of concentration c_s , is used to spike the flasks using a different volume, V_s , in each case. Normally, regularly increasing increments of V_s are used. Depending on the method of analysis other chemicals are added (complexing agents, buffer solutions, etc.). Finally each volumetric flask is filled to the calibration mark with the appropriate solvent. The concentration of the analyte in each of the volumetric flasks is

$$c_{\text{analyte}} = \frac{c_s V_s}{V_t} + \frac{c_x V_x}{V_t} \quad (1)$$

Using the standard additions method, within the linear calibration range, it is assumed that the response signal, R , of the analysis instrument is directly proportional to the analyte concentration; that is, $R = k c_{\text{analyte}}$. For each of the above flasks the appropriate equation is

$$R = k \left(\frac{c_s V_s}{V_t} + \frac{c_x V_x}{V_t} \right) = k \left[\frac{V_x}{V_t} \left(\frac{c_s V_s}{V_x} \right) + \frac{c_x V_x}{V_t} \right] \quad (2)$$

Defining c' as the increase in analyte concentration in the original sample volume (V_x) due to the added spikes gives

$$c' = \frac{c_s V_s}{V_x}$$

Equation 2 can be written in the form of a linear equation

$$R = m c' + b$$

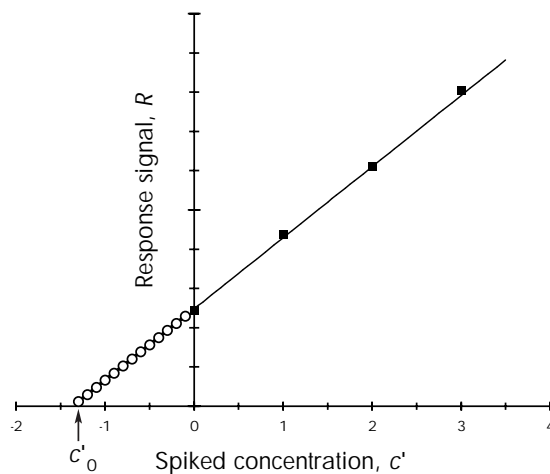


Figure 1. A representation of the standard additions method showing experimental data points (■) and the extrapolated linear regression line (o o o), which intercepts the x -axis at $c' = c'_0 = -c_x$.

with slope

$$m = k \frac{V_x}{V_t}$$

and intercept

$$b = k \frac{c_x V_x}{V_t}$$

which is shown graphically in Figure 1.

The x -axis intercept, c'_0 , of this extrapolated graph occurs at $R = 0$ and $b = -m c'_0$, which can be written as $b/m = -c'_0$. On substituting the above equations for the slope and the intercept, this simplifies to give $c_x = b/m = -c'_0$. Using this relationship the value of c_x may be obtained by either graphical extrapolation to c'_0 or by applying linear regression to the (R, c') data set to obtain the best-fit values for b and m (2, 3). The precision of c_x as expressed by the standard error, s_c , is then estimated from the error in c'_0 (Method 1) or from the errors in b and m (Method 2), as described in the next section.

Estimates of Precision

In the following, the coordinates (R, c') have been replaced by the more conventional notation (y, x) . There are N data points (y_i, x_i) on the standard addition calibration graph. Some basic statistical equations used in this paper are listed below.

$$\bar{x} = \frac{\sum x_i}{N} \quad (3)$$

$$\bar{y} = \frac{\sum y_i}{N} \quad (4)$$

$$S_{xx} = \sum (x_i - \bar{x})^2 = \sum x_i^2 - N(\bar{x})^2 \quad (5)$$

$$S_{yy} = \sum (y_i - \bar{y})^2 = \sum y_i^2 - N(\bar{y})^2 \quad (6)$$

$$S_{xy} = \sum (x_i - \bar{x})(y_i - \bar{y}) = \sum x_i y_i - N(\bar{x}\bar{y})^2 \quad (7)$$

$$s_y = \sqrt{\frac{S_{yy} - m^2 S_{xx}}{N - 2}} \quad (8)$$

$$m = \frac{S_{xy}}{S_{xx}} \quad (9)$$

$$b = \bar{y} - m\bar{x} \quad (10)$$

$$s_m = \frac{s_y}{\sqrt{S_{xx}}} \quad (11)$$

$$s_b = s_y \sqrt{\frac{\sum x_i^2}{NS_{xx}}} \quad (12)$$

Method 1. Extrapolation Method

As indicated previously, $c_x = -c_0'$. In the extrapolation method the appropriate equation (4, 5) for determining s_c , the standard error in c_x , is

$$s_c^2 = \frac{s_y^2}{m^2} \left(\frac{1}{N} + \frac{\bar{y}^2}{m^2 S_{xx}} \right) \quad (13)$$

where s_y is the standard deviation around the regression line, also known as the standard deviation of the residuals, and is given by eq 8.

Method 2. Algebraic Method

As indicated previously, $c_x = b/m$. Applying the law of propagation of errors (1, 6, 7), the relative variance in c_x is equal to the sum of the relative variances of b and m

$$\left(\frac{s_c}{c_x} \right)^2 = \left(\frac{s_b}{b} \right)^2 + \left(\frac{s_m}{m} \right)^2 \quad (14)$$

or

$$s_c^2 = c_x^2 \left(\frac{s_b}{b} \right)^2 + c_x^2 \left(\frac{s_m}{m} \right)^2 \quad (15)$$

where the standard deviations of the slope, s_m , and intercept, s_b , are given by eqs 11 and 12.

Examples

For these two methods, two examples will be used to compare the standard errors, s_c , and the associated 95% confidence limits for c_x . The first example is taken from a text (2). The second is from a student's analysis for lead using anodic stripping voltammetry in which the analyte matrix is unknown.

Example 1

A 10.0-mL (V_0) natural water sample was pipetted into each of five 50-mL (V_1) volumetric flasks. A spike of standard Fe^{3+} solution, 11.1 mg/L (c_0), was added to each flask using the following volumes: 0, 5.0, 10.0, 15.0, and 20.0 mL (V_2). Excess thiocyanate was added to the flasks to give the red complex $\text{Fe}(\text{SCN})^{2+}$. After dilution to volume the absorbances were measured at an appropriate analytical wavelength. The absorbance readings, R , for the five solutions were recorded as 0.240, 0.437, 0.621, 0.809, and 1.009, respectively.

Applying linear regression to the (R , c') data set gives $m = 0.03441$, $b = 0.2412$, and c_x (or $c_{\text{Fe}})$ = 7.01 mg/L. Table 1 gives the precision estimates as calculated by the two methods introduced above. Included is the 95% confidence limit for which $t = 3.182$ for $N - 2 = 3$ degrees of freedom. Concentrations are in mg/L.

Example 2

Twenty-five milliliters (V_x) of a lead-contaminated water sample, 25.0 mL of support electrolyte, and 1.0 mL of a 10.0 mg/L cadmium standard (an internal standard) were pipetted into a polarographic cell. Anodic stripping was performed on this solution and the ratio of the lead to cadmium stripping currents was measured to be 0.86. The solution was then spiked successively with 0.50, 1.00, 1.50, 2.0, and 2.50 mL (V_0) of 10.0 mg/L lead standard (c_0). After each addition the anodic stripping polarogram was recorded and the current ratio (R) was determined. The successive values of R were 1.11, 1.44, 1.74, 2.04, and 2.33.

Applying linear regression to the (R , c') data set gives $m = 1.491$, $b = 0.8410$, and c_x (or $c_{\text{Pb}})$ = 0.564 mg/L. Table 2 gives the precision estimates as calculated by the two methods introduced above. Included is the 95% confidence limit for which $t = 2.778$ for $N - 2 = 4$ degrees of freedom. Concentrations are in mg/L.

Table 1. Precision Estimates for Methods 1 and 2

| Statistical Measure | Method | |
|---|-------------|-------------|
| | 1 | 2 |
| Standard error, s_c | 0.159 | 0.123 |
| 95% confidence interval margin of error, ts_c | ± 0.51 | ± 0.39 |
| Relative margin of error, $100(ts_c/c_x)$ | $\pm 7.3\%$ | $\pm 5.6\%$ |

Table 2. Precision Estimates for Methods 1 and 2

| Statistical Measure | Method | |
|---|-------------|-------------|
| | 1 | 2 |
| Standard error, s_c | 0.0160 | 0.0119 |
| 95% confidence interval margin of error, ts_c | ± 0.045 | ± 0.033 |
| Relative margin of error, $100(ts_c/c_x)$ | $\pm 7.9\%$ | $\pm 5.9\%$ |

For examples 1 and 2, method 2 gives a lower estimate of the standard error. Can this be generalized? This is answered by comparing the two methods, eq 13 and eq 15, and using the statistical equations (3–12). For method 1, eq 13 may be rewritten as

$$s_c^2 = \frac{s_y^2}{m^2} \left(\frac{1}{N} + \frac{\bar{y}^2}{m^2 S_{xx}} \right) = \frac{s_y^2}{m^2 S_{xx}} \left(\frac{S_{xx}}{N} + \frac{\bar{y}^2}{m^2} \right) = \frac{s_y^2}{m^2 S_{xx}} \left[\frac{S_{xx}}{N} + \frac{(b + m\bar{x})^2}{m^2} \right]$$

Substituting for S_{xx} from eq 5 and rearranging gives

$$s_c^2 = \frac{s_y^2}{m^2 S_{xx}} \left(\frac{\sum x_i^2}{N} + \frac{b^2}{m^2} + \frac{2b\bar{x}}{m} \right) \quad (16)$$

For method 2, substituting $c_x = b/m$ into eq 15 gives

$$s_c^2 = \frac{b^2}{m^2} \left[\left(\frac{s_b}{b} \right)^2 + \left(\frac{s_m}{m} \right)^2 \right] = \frac{s_b^2}{m^2} + \frac{b^2 s_m^2}{m^4} \quad (17)$$

Using eqs 11 and 12 (defining s_m and s_b), eq 17 becomes

$$s_c^2 = \frac{s_y^2 \sum x_i^2}{m^2 N S_{xx}} + \frac{b^2 s_y^2}{m^4 S_{xx}} = \frac{s_y^2}{m^2 S_{xx}} \left(\frac{\sum x_i^2}{N} + \frac{b^2}{m^2} \right) \quad (18)$$

On comparing the s_c^2 terms for the two methods, eqs 16 and 18, one finds that the term

$$\frac{2b\bar{x}s_y^2}{m^3 S_{xx}}$$

is missing from the latter equation. For a standard additions analysis this term is positive. Therefore s_c obtained using the extrapolation method equation will always be greater than s_c obtained using the algebraic method.

The following shows that the apparent difference is the result of an erroneous assumption in the derivation of s_c in eq 14 (1, 2). The missing term can be explained once the law of propagation of errors is applied properly.

A better approximation for

$$\left(\frac{s_c}{c_x} \right)^2$$

is obtained by including the covariance, s_{mb} , between the estimates of the slope and the y -axis intercept, as is shown in eq 19 (8).

$$\left(\frac{s_c}{c_x} \right)^2 = \left(\frac{s_m}{m} \right)^2 + \left(\frac{s_b}{b} \right)^2 - 2 \left(\frac{s_{mb}}{mb} \right) \quad (19)$$

or

$$s_c^2 = c_x^2 \left(\frac{s_m}{m} \right)^2 + c_x^2 \left(\frac{s_b}{b} \right)^2 - 2c_x^2 \left(\frac{s_{mb}}{mb} \right) \quad (20)$$

where

$$s_{mb} = -\frac{\bar{x}s_y^2}{S_{xx}}$$

It should be noted that the covariance s_{mb} need not diminish with increasing the number of measurements.

Compared to eq 15, eq 20 contains the extra term

$$\frac{2c_x^2 s_{mb}}{mb}$$

and

$$-\frac{2c_x^2 s_{mb}}{mb} = -\frac{2bs_{mb}}{m^3} = -\frac{2b}{m^3} \left(-\frac{\bar{x}s_y^2}{S_{xx}} \right) = \frac{2b\bar{x}s_y^2}{m^3 S_{xx}}$$

which is identical to the missing term in s_c^2 given by method 2.

Conclusion

In a standard additions analysis the slope and intercept are generated from one data set and are not determined independently; hence the covariance term is significant. As recently pointed out (9), this erroneous assumption of independence of slope and intercept estimates has led to incorrect determinations of standard errors in other applications. Consequently, it is recommended that the extrapolation method equation is used when estimating the precision of standard additions analyses.

Acknowledgments

We would like to thank two referees for their helpful comments. G. R. Bruce worked on this project while on an extended study leave from the Okanagan University College. The work of P. S. Gill was partially supported by a grant from the Natural Sciences and Engineering Research Council of Canada.

Literature Cited

- Bader, M. *J. Chem. Educ.* **1980**, *57*, 703–706.
- Skoog, D. A.; Leary, J. J.; Nieman, T. A. *Principles of Instrumental Analysis*, 5th ed.; Harcourt Brace: Orlando, FL, 1998; 15–18.
- Meir, P. C.; Zund, R. E. *Statistical Methods in Analytical Chemistry*; Wiley: New York, 1993; pp 84–120.
- Draper, N. R.; Smith, H. *Applied Regression Analysis*, 2nd ed.; Wiley: New York, 1981; pp 8–55.
- Miller, J. C.; Miller, J. N. *Statistics for Analytical Chemistry*, 3rd ed.; Ellis Horwood: Chichester, UK, 1993; pp 101–139.
- Skoog, D. A.; Leary, J. J.; Nieman, T. A. *Op. cit.*; pp A15–A16.
- Moritz, P. *Comprehensive Analytical Chemistry*, Vol. XI; Elsevier: New York, 1981; pp 67–109.
- Rice, J. A. *Mathematical Statistics and Data Analysis*, 2nd ed.; Duxbury: Belmont, CA, 1995; pp 149–154, 514.
- Meyer, E. F. *J. Chem. Educ.* **1997**, *74*, 1339–1340.