Short communication

Effect of multiple error sources on the calibration uncertainty

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ABSTRACT

The calibration uncertainty associated with the determination of metals at trace levels in a drinking water sample by ICP-MS was estimated when signals were affected by two error contributions, namely instrumental errors and operational condition errors. The calibration uncertainty was studied by using J concentration levels measured I times, as usual in experimental calibration procedures. The instrumental error was random in character whilst the operational error was assumed systematic at each concentration level but random among the J levels. The presence or the absence of the two error contributions was determined with an F-test between the ordinary least squares residual variance of the mean responses at each concentration and a pooled variance of the replicates. The theory was applied to the calibration of 30 elements present in a multi-standard solution and then to the analysis of boron, calcium, lithium, barium and manganese in a real drinking water sample. The need of using the proposed approach as calibration for almost all the analyzed elements resulted evident. The presence or the absence of the two error contributions was determined with an F-test between the ordinary least squares residual variance of the mean responses at each concentration and a pooled variance of the replicates. It was found that in the former instance the uncertainty determined using a two-components variance regression was greater than that obtainable from the one-variance regression.

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1. Introduction

The estimation of the uncertainty associated with the calibration is one of the most cumbersome issues encountered both in method validation and in the determination of an unknown concentration. In fact, operational variations may occur caused either when several matrices are analyzed for the same analyte or in prolonged analytical procedures or in the presence of significant change in the temperature (Ellison, Barwick, & Farrant, 2009; Juelicher, Gowik, & Uhlig, 1998; Juelicher, Gowik, & Uhlig, 1999; Lavagnini, Fedrizzi, Versini, & Magno, 2009). Typical examples of these cases are the analysis of a doping substance searched in various animal tissues and the need of a re-calibration procedure in routine analysis prolonged for long time. These facts imply that at least two contributions of error variances may be present namely, the instrumental and the operational ones. In these cases the regression is faced with a two-component variance model (Juelicher et al., 1998; Muller & Uhlig, 2001; Searle, Casella, & McCulloch, 1992). Anyway, the uncertainty of the calibration curve is usually calculated by means of a one-variance model, the instrumental one, accounting for the total number of measurements used in the construction of the curve, the number of replicates from each solution, and the concentration levels. In this experimental design, the number of measurements came from instrumental runs and the ith replicate from the jth solution is a function of the concentration level, x_j, given by $y_{ij} = \beta_0 + \beta_1 x_j + \varepsilon_{ij}$, where ε_{ij} is the instrumental error. This approach was recently used for metals ICP-MS determination in food samples at trace level (Yenisoy-Karakaş, 2012). When working at very low concentration levels, however, a lot of non-instrumental error sources become relevant and may dramatically influence the analytical result requiring the two-components variance approach for the regression procedure. In fact, sample preparation, anthropic and environmental pollution, standard reagent purity, glassware quality, become more and more important and may make the non-instrumental error variance very significant. Consequently, if J solutions are prepared and then measured I times, the calibration line construction must be faced with a different approach as an additive systematic contribution affects the I replicates at the jth concentration level. This systematic error is random in character among the J concentration levels.

This paper is aimed at studying the estimation of the uncertainty of the calibration curve when the relationship between response and concentration is given by $y_{ij} = \beta_0 + \beta_1 x_j + \eta_j + \varepsilon_{ij}$, where η_j represents the non-instrumental error. The approach represents an application of the random model ANOVA procedure to

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the calibration responses in the presence of the instrumental error and additional error sources (Searle et al., 1992). The model will be applied to synthetic data, generated using the Monte Carlo method, in order to give the details of uncertainty calculation of the concentration associated with the calibration curve uncertainty. Further, the developed model will be used to determine boron, calcium, lithium, barium and manganese in real drinking water samples. Metals like boron, manganese and barium were chosen as they are present at trace levels and subjected to legal limits, whereas calcium and lithium are usually present at higher concentrations with no legal limits. Testing on the ICP-MS technique is particularly suitable owing to the intrinsic feature of the output signals which are integrals of many repeated measurements in time. The signal is usually considered as a single value but it is actually a mean value with a proper variance.

2. Materials and methods

2.1. Drinking water samples

The drinking water samples were obtained from "Fonte Margherita" (Torrebelvicino, Vicenza, Italy). Samples were added of 3% (v/v) nitric acid to uniform the solutions to the calibration ones.

2.2. Reagents and instrumentation

All reagents were of analytical grade and were used as purchased: 69% HNO_3 (CAS: 7697-37-2) (ARISTAR®), multi-element standard solution CPAchem (10 mg L^{-1}) ICP-MS calibration standard Ref. N: MS19EB.10.2N.L1. All solutions were prepared in milliQ ultrapure water obtained with a Millipore Plus System (Milan, Italy, resistivity 18.2 Mohm cm^{-1}). The ICP-MS was tuned daily using a 1 μg L^{-1} tuning solution containing ^{140}Ce, ^{7}Li, ^{205}Tl and ^{89}Y (Agilent Technologies, UK). A 100 μg L^{-1} solution of ^{45}Sc and ^{115}In (Aristar®, BDH, UK) prepared in 3% (v/v) nitric acid was used as an internal standard through addition to the sample solution via a *T*-junction.

All the elements were measured by using inductively coupled plasma coupled to a mass spectrometer (ICP-MS) Agilent Technologies 7700× ICP-MS system (Agilent Technologies International, Japan, Ltd., Tokyo, Japan). The MS detector is equipped with an octupole collision cell operating in kinetic energy discrimination mode for the removal of polyatomic interferences and argon-based interferences. The instrument was optimized daily to achieve optimum sensitivity and stability according to manufacturer recommendations. Typical operating conditions and data acquisition parameters are summarized in Table 1.

2.3. Procedures

Multielement standard solutions were prepared in 3% v/v HNO_3. The calibration solutions were prepared by gravimetric serial dilution from multi-element standard solutions, at six different concentrations (1 ng L^{-1}–100 mg L^{-1}). Calibration plots were obtained with an internal standard. Blank samples of ultrapure water and reagents were also prepared using the same procedures adopted for the samples. All blank levels obtained were appropriately subtracted.

2.4. Statistical model

When replicated measurements on the same standard at the concentration level x_j are made, a general relationship including the *y* measurement, the *x* concentration, and the analytical conditions at the spiked level *j* may be expressed in the form

Table 1
Instrumental operative conditions for ICP-MS.

Instrument	Agilent 7700× ICP-MS
RF power	1550 W
RF matching	1.8 V
Plasma gas flow rate	15 L min^{-1} Ar
Auxiliary gas flow rate	1.0 L min^{-1} Ar
Carrier gas flow rate	1.05 L min^{-1} Ar
Make-up gas flow rate	0.0 L min^{-1} Ar
He gas flow	4.3 mL min^{-1}
CeO$^+$/Ce$^+$	0.902%
Ratio(2+) 70/140	0.944%
Nebuliser	Microflow PFA nebuliser
Spray chamber	Scott double-pass type at 2 °C
Torch	Quartz glass torch
Sample uptake rate	0.1 mL min^{-1}
Sample cone Nickel	1.0 mm aperture i.d.
Skimmer cone nickel	0.5 mm aperture i.d.
Sampling depth	8.5 mm
Detector mode	Dual (pulse and analog counting)
Dwell time/mass	1000 ms
Replicate	9

$$y_{ij} = \beta_0 + \beta_1 x_j + \eta_j + \varepsilon_{ij} \tag{1}$$

where $i = 1, 2, \ldots, I$ denotes the measurements at the level x_j. The instrumental error, ε_{ij}, is assumed to have a normal distribution with zero mean and constant variance $\sigma_\varepsilon^2 : \varepsilon_{ij} \sim N(0, \sigma_\varepsilon^2)$. The term η_j takes into account random effects due to operating conditions in preparing the *j*th standard and assumed constant among the *I* replications at x_j. It is assumed normally distributed with zero mean and variance $\sigma_\eta^2 : \eta_j \sim N(0, \sigma_\eta^2)$. Moreover, the random variables ε_{ij} and η_j are assumed uncorrelated. Therefore the random variable y_{ij} is also normally distributed with two components of variance, that is $\sigma_y^2 = \sigma_\eta^2 + \sigma_\varepsilon^2$, and $y_{ij} \sim N(\beta_0 + \beta_1 x_j, \sigma_\eta^2 + \sigma_\varepsilon^2)$. Eq. (1) corresponds to the random effects ANOVA model in regression (Searle et al., 1992).

2.4.1. Statistical analysis of the calibration experiment

The β_0 and β_1 parameters in Eq. (1) may be estimated by the ordinary least squares method (OLS) referring to the mean response at each concentration level, $\bar{y}_j = \beta_0 + \beta_1 x_j + \eta_j + \bar{\varepsilon}_j$, where $\bar{\varepsilon}_j = \sum_i^I \varepsilon_{ij}/I \sim N\left(0, \frac{\sigma_\varepsilon^2}{I}\right)$ (Searle et al., 1992). Since the sum $\gamma_j = \eta_j + \bar{\varepsilon}_j$ is also normally distributed, $\gamma_j \sim N\left(0, \sigma_\eta^2 + \frac{\sigma_\varepsilon^2}{I}\right)$, the average signal $\bar{y}_j = (1/I)\sum_{i=1}^I y_{ij}$ at the concentration level x_j is also Gaussian, $\bar{y}_j \sim N(\beta_0 + \beta_1 x_j, \sigma_{\bar{y}_j}^2)$ where $\sigma_{\bar{y}_j}^2 = \sigma_{\gamma_j}^2$ is given by

$$\sigma_{\bar{y}_j}^2 = \sigma_\eta^2 + \frac{\sigma_\varepsilon^2}{I} \tag{2}$$

The OLS estimates of the parameters β_0 and β_1 are obtained minimizing the sum $\sum_j^J \gamma_j^2$, and are given by $\hat{\beta}_0 = \bar{y} - \hat{\beta}_1 \bar{x}$, $\hat{\beta}_1 = \frac{\sum_{j=1}^J (x_j - \bar{x})\bar{y}_j}{S_{xx}}$, where $\bar{y} = (1/J)\sum_{j=1}^J \bar{y}_j, \bar{x} = (1/J)\sum_{j=1}^J x_j$, and $S_{xx} = \sum_{j=1}^J (x_j - \bar{x})^2$. The OLS estimate of the variance $\sigma_{\bar{y}_j}^2$ is given by the residual variance

$$\hat{\sigma}_{\bar{y}_j}^2 = \frac{\sum_{j=1}^J (\bar{y}_j - \hat{y}_j)^2}{J - 2} \tag{3}$$

2.4.2. Evidence of the experimental presence of the variance σ_η^2

The significant presence of additional errors may be checked with an *F*-test (Ellison et al., 2009). From Eqs. (2) and (3) the statistic