Letters

Letter by Dr. M. J. Cardone

I have read the recent paper by Bosch Reig and Campíns Falcó[1] and would like the opportunity to show that it actually represents an innovative extension of the corrigible error correction (CEC) technique.[2,3] They have shown that with their procedure an analyte concentration can be obtained free of bias error (both constant and proportional) both from a direct known interferent and from the true method blank under carefully stipulated conditions. Further, a determination of an interferent concentration can also be made simultaneously. It is this latter part of the procedure that I would also like to critique.

x,y-Co-ordinates Coincident Point

The extension of the CEC technique lies in the use of multipoint signal data (in this instance, a spectrum) such as is provided with spectrophotometry. By use of standard additions plots at each of two carefully chosen wavelengths (validated as described in reference 1) Bosch Reig and Campíns Falcó utilise the condition of remote extrapolation to the *x,y*-co-ordinates coincident point in the same manner as was carried out in a recent CEC paper.[4] This procedure yields an analyte concentration in the units of the abscissa (*x*-co-ordinate) free from both constant and proportional systematic errors. (For the significance of the *y*-co-ordinate see later.)

Why the analyte concentration would be bias free, a most remarkable fact, is not discussed in the literature as far as I am aware, was not fully explained in reference 4 nor was it explained in reference 1.

Proportional error is eliminated by virtue of the fact that the method of standard additions (MOSA) provides an *in situ* normalisation because the same procedural operation is performed on the unspiked and spiked samples, so that a constant percentage bias is introduced on the samples across the dynamic range.[2,3]

How the constant error is eliminated is to be found in the mathematical operation for the calculation of the coincident point, the intersection of the two standard additions plots. Solving two straight-line equations simultaneously reduces to a subtraction of the two *y*-response signals, thereby resulting in a cancellation of the constant error that is incorporated in each. Examples of this principle can be found in the classical Youden two-sample collaborative interlaboratory procedure that yields the well known Youden plot[5,6] and in the first quantitative calculation of the true method blank by Kimball and Tufts,[7] described more fully in reference 3.

The ability to obtain a bias-free analyte concentration is then not unique as it can be achieved via the CEC technique from two standard additions plots using two different sized samples for spiking.[4] However, the ability presented by Bosch Reig and Campíns Falcó[1] to separate constant error into its components whenever multipoint data permits is a significant development deserving of close examination.

y-*Co-ordinate coincident point*

Bosch Reig and Campíns Falcó state that the *y*-co-ordinate, A_H (Fig. 1 of reference 1) is the analytical signal due to the interferent. This statement is only partially correct. In fact, the A_H signal is due to the interferent plus the true method blank, or TYB (total Youden blank) as it is called in CEC terminology. It should be noted that both of these components are constant systematic errors with the former, the interferent, of the fixed systematic type, hence corrigible as against variable systematic errors that are incorrigible.[2,3] Bosch Reig and Campíns Falcó carefully and correctly ensure that the interferent is a constant corrigible error by their stipulation that it exhibits constant absorbance over the wavelength range of interest (Fig. 3 of reference 1) and that this stipulation is validated (Fig. 5 of reference 1).

In Fig. 1 of reference 1, the analytical signal, S, consists of that due to the analyte, the interferent and the true method blank, the TYB. The overlooking of the TYB component has been a consistent error of omission in the analytical literature and which has been discussed thoroughly in the CEC literature.[2,3] Failure to subtract the TYB component from the total signal is an incorrect standard additions model, as was shown recently,[4] and produces biased results. Notwithstanding, the procedure of Bosch Reig and Campíns Falcó results in a separation of the analyte concentration from the total constant error concentration and this fact is of considerable importance.

Why then do Bosch Reig and Campíns Falcó obtain such acceptable results for the interferent concentration in spite of the fact that it is the total constant error concentration and not just the interferent concentration? As I do not have the raw data, I cannot answer this question conclusively. However, I do have an explanation that they can examine. As the samples used were simple solution dye mixtures, essentially matrixless in the sense that they are indistinguishable from reference standard mixtures for a reference standard curve, and as in spectrophotometry, it is usual for Beer's law plots to pass through the origin, it is highly probable that the TYB component for these examples is statistically essentially zero. Any small acceptable analytical value would be contained within the normal variance of the method.

Recommended CEC Procedure

Bosch Reig and Campíns Falcó have presented their procedure as a free-standing MOSA technique, independent of a standard response curve. This is most certainly true when applicable under the stipulated conditions, for the bias-free determination of the analyte concentration. However, if the method were to be applied to more complex materials, such as pharmaceutical dosage forms or environmental samples where a significant TYB value were present, the determination of a known interferent would not be possible without bias from the TYB. In such a situation, correction of the analytical signal, S, for use in Fig. 1 of reference 1 would be required. An example of applying the TYB for total signal correction for use in a free-standing MOSA application was given recently by Ferrus and Torrades.[8]

Unless Bosch Reig and Campíns Falcó present some new information in a subsequent paper, the only way in which a true sample blank can be determined at present is by means of the Youden one-sample regression procedure[2,3,8] or its earlier equivalent alternatives.[7] In the examples of reference 1, this would simply involve selection of an appropriate dye mixture sample and varying the amount over the linear dynamic range across and including the maximum response values of the standard additions samples, all at one of the selected wavelengths. The resulting intercept value of the linear regression straight line is the TYB. It should be noted that in this procedure, the TYB value represents a separation from a total signal in which the analyte and interferent signal components remain superimposed. Under the conditions stipulated for the wavelengths, the probabilities are that the TYB determined in a similar manner at the second wavelength would be a statistically equivalent value. If they are not, a complex matrix interaction problem would be indicated, and a question of validity of the subsequent interferent determination would have to be examined.

In conclusion, Bosch Reig and Campíns Falcó have presented a valid and important addition to the free-standing

status of the method of standard additions for a bias-free analyte determination with their multipoint data technique, where applicable. However, the simultaneous determination of a corrigible interferent is only possible if a true method blank, TYB is absent. If present, a Youden one-sample regression determination of the TYB is required in order to correct the total analytical signals. It is for this reason, as this is the situation more often than not with complex samples, that I consider their technique to be an important and potentially useful extension of the CEC technique.

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Reply by Dr. F. Bosch Reig and Dr. P. Campíns Falcó

First, we thank Dr. M. J. Cardone for the interest he has taken in our paper.[1] We know his work and consider it to be a major contribution to the obtainment of unbiased analytical results.

His letter includes some remarks about the determination of the interferent by our method, which we will deal with here with the aim of extending and clarifying the fundamentals of the method.

We originally conceived and developed the H-point standard additions method (HPSAM) for application to multicomponent samples and samples with complex matrices. In reference 1 the method was applied to matrixless samples such as mixtures of dyes with the aim of showing how to calculate an analyte concentration ($C_X = -C_H$, abscissa of the H-point) free from constant and proportional systematic errors.

In dealing with binary mixtures, the HPSAM also allows the interferent concentration (C_Y) to be determined from A_H (the ordinate of the H-point) in the absence of a constant bias—only the proportional bias is eliminated by the HPSAM in quantifying interferents.

The occurrence of a constant bias due to the sample matrix (TYB, total Youden blank) was always considered in developing our method. The samples used in reference 1 were mixtures of dyes, and the statistically calculated TYB was virtually zero. This is also frequently the situation with more complex samples than those used in reference 1. Moreover, when the analyte signal is only modified by the matrix effect, the HPSAM allows both proportional and constant bias errors to be corrected, and the latter to be calculated. In this event, the two selected wavelengths can be chosen arbitrarily thanks to the very nature of the TYB, and an unbiased analyte concentration can be obtained by performing a single set of experiments and applying the multipoint data technique. Application of the corrigible error correction (CEC) technique,[2,3] on the other hand, entails carrying out two sets of experiments.

The above considerations are expressed theoretically below. Let us consider an unknown containing an analyte X and an interferent Y subject to a constant bias error, TYB. As previously,[1] we select two wavelengths (λ_1, λ_n) from the analyte and the interferent spectra and relate them with their corresponding absorbance values through the following expressions:

$$A_i = b_i + m_i\lambda_j \qquad (\lambda_1 \geqslant \lambda_j \geqslant \lambda_n) \quad .. \quad (1)$$

for the analyte (X) and

$$A' = b + m\lambda_j \qquad (m = 0) \quad .. \quad .. \quad (2)$$

for the interferent (Y), where b and m are the intercepts and slopes of the corresponding straight lines, respectively. The subscript i refers to the different solutions for m additions of the analyte concentration prepared in order to apply the standard additions method and the subscript j indicates a wavelength in the λ_1–λ_n range.

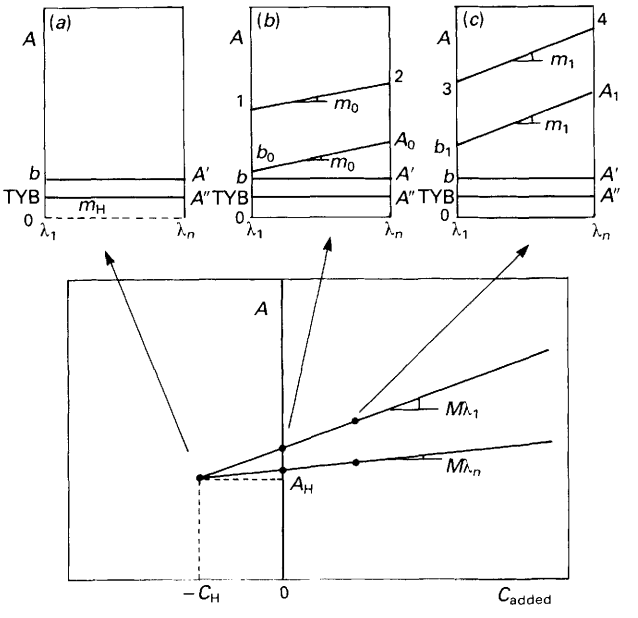

Fig. 1. H-point standard additions plots. For details see text

This condition is fulfilled because the interferent features a constant absorbance at the two selected wavelengths.

Let us also consider the TYB, which will normally be the same at the two selected wavelengths, *i.e.*,

$$A'' = \text{TYB} \quad .. \quad .. \quad .. \quad .. \quad (3)$$

Hence, the over-all equation corresponding to the first solution ($i = 0$) used to apply the method of standard additions (MOSA), which only contains the sample, with no added analyte, will be

$$A_0 + A' + A'' = b_0 + b + m_0\lambda_j + \text{TYB} \quad .. \quad (4)$$

The plot of A *versus* λ is shown in Fig. 1(*b*).

The MOSA starts by calculating the unknown concentration by extrapolation when the ordinate value is zero. Then, taking into account that, according to the HPSAM, $A_i = 0$ for $m_i = m = 0$ this is the H-point [Fig. 1(*a*)], at which the slope of the straight line of the $A = f(\lambda)$ plot for the analyte equals that of the interferent. The over-all equation will thus be reduced to

$$b + \text{TYB} = A' + A'' = A_H \quad .. \quad .. \quad (5)$$

Obviously, for TYB = 0, $b = A' = A_H$ and therefore, b can be related to the interferent concentration. Hence, in the absence of the interferent, TYB = $A'' = A_H$ and TYB can be determined readily.

Let us now consider the instance involving two constant systematic errors. The $A = f(\lambda)$ plot, corresponding to the first addition of the MOSA, is shown in Fig. 1(*c*) and the over-all equation is

$$A_1 + A' + A'' = b_1 + b + \text{TYB} + m_1\lambda_j \quad .. \quad (6)$$

These concepts can be clarified by plotting the absorbance as a function of the added analyte concentration (C_i) at the two selected wavelengths (Fig. 1). The MOSA equation will be

$$A\lambda_1 = b_i + b + M\lambda_1 C_i + \text{TYB} \quad .. \quad .. \quad (7)$$

for λ_1, and

$$A\lambda_n = A_i + A' + M\lambda_n C_i + \text{TYB} \quad .. \quad .. \quad (8)$$

for λ_n, where $M\lambda_1$ and $M\lambda_n$ are the slopes of the MOSA plots.

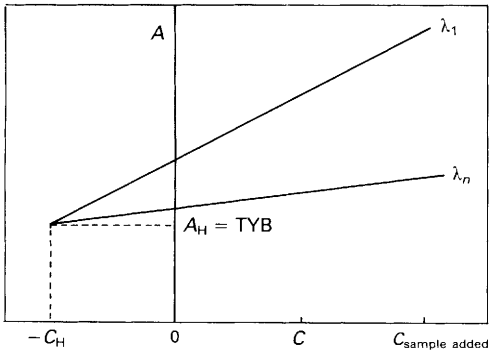

Fig. 2. Plot of the H-point standard additions method

The H-point is common to the two MOSA plots and is given by

$$b_i + b + M\lambda_1 C_i + \text{TYB} = A_i + A' + M\lambda_n C_i + \text{TYB} \quad (9)$$

or

$$C_i(M\lambda_1 - M\lambda_n) = A_i + A' - b_i - b \quad (10)$$

As $A' = b$, then

$$C_i = (A_i - b_i)/(M\lambda_1 - M\lambda_n) = C_X(M\lambda_n - M\lambda_1)/(M\lambda_1 - M\lambda_n) = -C_X = -C_H \quad (11)$$

The analyte concentration can thus be obtained free from constant and proportional bias errors.

By substituting the C_i value obtained into equations (7) and (8) A_H can be determined through equation (12)

$$A_H = b_i + b + M\lambda_1(-C_H) + \text{TYB} \quad (12)$$

As $b_i = M\lambda_1 C_H$, then

$$A_H = b + \text{TYB} \quad (13)$$

Hence, it is possible to obtain an unbiased analyte concentration directly on application of the HPSAM if the constant bias is the same at the two selected wavelengths.

The interferent concentration will be free from proportional, but not from constant bias, which requires evaluation by Youden's method[9] or the CEC technique.[2,3] We agree with Dr. M. J. Cardone on this point; however, it is not essential that the TYB be the same at the two selected wavelengths as it can be evaluated at both and later used to construct the HPSAM graph by plotting $A\lambda_1 - \text{TYB}\lambda_1$ and $A\lambda_n - \text{TYB}\lambda_n$ as a function of the added concentration.

The HPSAM was applied to mixtures of dyes and metal ions, and to organic compounds in reference 1. All samples were found to have a TYB of zero. However, we are currently dealing with multi-component mixtures and samples with complex matrices, which therefore require correction of the constant bias by applying the Youden method or the CEC technique, or even the HPSAM by substituting the added analyte concentration by the added sample concentration (Fig. 2). The A_H value thus obtained is equivalent to the TYB if this is equivalent at the two selected wavelengths. In this way, the HPSAM, like the Youden method and the CEC technique, permits the occurrence of TYB to be detected; in addition, it allows one to determine whether it is constant at the two selected wavelengths.

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References

1. Bosch Reig, F., and Campíns Falcó, P., *Analyst*, 1988, **113**, 1011.
2. Cardone, M. J., *Anal. Chem.*, 1986, **58**, 438.
3. Cardone, M. J., *J. Assoc. Off. Anal. Chem.*, 1983, **66**, 1283.
4. Cardone, M. J., *Anal. Chem.*, 1987, **59**, 2818.
5. Youden, W. J., and Steiner, E. H., "Statistical Manual of the AOAC," Association of Official Analytical Chemists, Arlington, VA, 1975.
6. Cardone, M. J., *J. Assoc. Off. Anal. Chem.*, 1983, **66**, 1257.
7. Kimball, R. H., and Tufts, L. E., *Anal. Chem.*, 1947, **19**, 150.
8. Ferrus, R., and Torrades, F., *Anal. Chem.*, 1988, **60**, 1281.
9. Youden, W. J., *Anal. Chem.*, 1947, **19**, 946.