Send Orders of Reprints at reprints@benthamscience.net

20 *The Open Toxicology Journal,* 2013, *6,* (Suppl 1, M3) 20-26

Open Access

A Practical Guide to the Calculation of Uncertainty of Measurement

Mario Zilli*

ARPA F.V.G., Settore Laboratorio Unico, Laboratorio di Gorizia, Italy

Abstract: This paper would provide the analyst with an easy to use step-by-step guide to calculate the uncertainty of measurement in implementing a new analytical method. There are two main ways to attain such an achievement. The first is to consider all the possible sources of variability and then to sum up all of them in the final calculation. The second is taking part in a collaborative trial and processing the resulting statistics. Both methods imply different advantages and drawbacks. It is up the analyst to choose the one fits best his requirements. Anyway it is nearly inescapable providing, along with the test result, its own uncertainty.

Keywords: Uncertainty, combined standard uncertainty, expanded uncertainty, coverage factor.

1. INTRODUCTION

Setting aside what is prescribed by any national legislation, it is clear by now that the Forensic Toxicologists must be concerned with the quality of the results they produce. In this regard, the use of qualified personnel, suitable instrumentations and highly reliable analytical methods is necessary.

This entire subject is stated by a standard well known to those who practise the profession of a chemical analyst, which is the UNI CEI EN ISO/IEC 17025:2005. This standard has been imposed by the European Community for matters concerning food and environmental surveillance, but it has also become part of the guidelines of several National Associations of Clinical Toxicology. This standard pledges the analyst to use validated analytical methods, issued by one of the national or international Standards Organizations or, at least, by an internationally acknowledged Scientific Society.

In this paper we aim to describe a particular aspect of the validation process, maybe the most important one: the uncertainty of measurement. It is clear in fact that nowadays the Clinical or Forensic Toxicologists cannot just supply a mere number as the result of their analytical work. They must provide, along with such a number, a range stating which the probability is that the true value – or the one accepted as such – lies within that range. This is the best that statistics can achieve from such a result. It is clear that the better the analytical performance quality is, the narrower that range will be, and the closer the result will approach the true value: all that, of course, within a previously selected level of probability – in other words at the selected confidence level. It is also obvious that the uncertainty assessment of a method, acquired in the validation process phase, involves additional hard work, but the ensuing output is worthwhile in terms of scientific value and, not less important, it is reliable from any point of view.

*Address correspondence to this author at the ARPA F.V.G., Settore Laboratorio Unico, Laboratorio di Gorizia, Italy; Tel: +390481583800; Fax: +390481583814; E-mail: mario.zilli@arpa.fvg.it

The uncertainty assessment can concern systematic errors, since the adjustment due to applying the method in question to a certified standard material (whose nominal value is accepted as the true value) undergoes a certain degree of dispersion in itself. This assessment though concerns in particular the dispersion of random errors which occurs when repeated tests are performed on the same sample, and it takes into account all the possible factors that can affect it. It should be pointed out that the awareness of such variability does not reduce, but instead it improves the knowledge about the measured value.

It follows that the uncertainty does not represent the difference between the measured and the true value (which will always be unknown), but it is an estimated interval that includes all the possible results of the measurement – including the true value – at the chosen confidence level.

Now let us give some definitions.

2. DEFINITIONS

Measurand: "Quantity intended to be measured" [ISO/IEC Guide 99:2007, 2.3].

Repeatability conditions: "Conditions where independent test results are obtained by the same method, on identical test items, in the same laboratory, by the same operator, using the same equipment within short time intervals" [ISO 5725-1:1994].

Intermediate repeatability conditions: Conditions where one or more of the previous variables can be changed (except the sample and the laboratory) (see ISO 5725-3:1994).

Reproducibility conditions: "Conditions where the results are obtained with the same method, on identical test items, in different laboratories, with different operators, with using different equipment" [ISO 5725-1:1994].

Confidence/significance level (α): "Maximum probability of rejecting the null hypothesis when in fact is true" [ISO 3534-1:2006].

1874-3404/13

Confidence interval: "Interval estimator (T_0,T_1) for the parameter θ with the statistics T_0 and T_1 as interval limits and for which it holds that $P[T_0 < \theta < T_1] \geq (1 - \alpha)$" [ISO 3534-1:2006].

Confidence interval and level: "Confidence interval gives an estimated range of values which is likely to include an unknown population parameter, the estimated range being calculated from a given set of sample data. If independent samples are taken repeatedly from the same population, and a confidence interval calculated for each sample, then a certain percentage (confidence level) of the intervals will include the unknown population parameter" [1].

Random (measurement) error: "Component of a measurement error that in replicate measurements varies in an unpredictable manner" [ISO/IEC Guide 99:2007, 2.19].

Systematic (measurement) error: "Component of a measurement error that in replicate measurements remains constant or varies in a predictable manner" [ISO/IEC Guide 99:2007, 2.17].

Precision: "The closeness of agreement between independent test results obtained under stipulated conditions" [ISO 5725-1:1994].

Repeatability: "Precision under repeatability conditions" [ISO 5725-1:1994].

Repeatability standard deviation: "The standard deviation of test results obtained under repeatability conditions" [ISO 5725-1:1994].

Reproducibility: "Precision under reproducibility conditions" [ISO 5725-1:1994].

Reproducibility standard deviation: "The standard deviation of test results obtained under repeatability conditions" [ISO 5725-1:1994].

Note: in any case the standard deviation is the positive square root of the variance.

Uncertainty (of measurement): a. "Non-negative parameter, characterizing the dispersion of the quantity values being attributed to a measurand, based on the information used" [ISO/IEC Guide 99:2007 – 2.26]. b. "Parameter, associated with the result of a measurement, that characterizes the dispersion of the values that could reasonably be attributed to the measurand" [ISO 21748:2010 – 2.14].

Note: The uncertainty generally includes many components which may be evaluated from experimental standard deviations based on repeated observations (Type A evaluation) or by standard deviations evaluated from assumed probability distributions based on experience or other information (Type B evaluation).

Law of propagation of uncertainty: "the uncertainty σ_z of a quantity $z = f(w_1, w_2, \ldots, w_N)$ that depends on N input quantities $w_1, w_2, \ldots, w_N$ is found from

$$\sigma_z^2 = \sum_{i-1}^{N}\left(\frac{\partial f}{\partial w_i}\right) \quad \sigma_i^2 + 2\sum_{i-1}^{N-1}\sum_{j-1+1}^{N}\frac{\partial f}{\partial w_i}\frac{\partial f}{\partial w_j}\sigma_i\sigma_j\rho_{ij}$$

where σ_i^2 is the variance of w_i and ρ_{ij} is the correlation coefficient of the covariance of w_i and w_j. If the input quantities are independent (as is often the case), then the covariance is zero and the second term of the above equation vanishes. The above equation is traditionally called the 'general law of error propagation', but this equation actually shows how the uncertainties (not the errors) of the input quantities combine [ISO, 46]; [2, 3].

Note: In all of the formulas listed below the assumption of the independence of the quantities to be summed up is made.

Combined Standard Uncertainty: "Standard uncertainty of the result of a measurement when that result is obtained from the values of a number of other quantities, equal to the positive square root of a sum of terms, the terms being the variances or covariances of these other quantities weighed according to how the measurement result varies with changes in these quantities" [ISO 21748:2010 – 2.2].

Coverage factor: "Numerical factor used as a multiplier of the combined standard uncertainty in order to obtain an expanded uncertainty" [ISO 21748:2010 – 2.2].

Expanded uncertainty: "Quantity defining an interval about a result of a measurement expected to encompass a large fraction of values that could be reasonably be attributed to the measurand" [ISO 21748:2010 – 2.2].

Certified Reference Material (CRM): reference material accompanied by documentation issued by an authoritative body and providing one or more specified property values with associated uncertainties and traceabilities, using valid procedures [ISO/IEC Guide 99:2007, 5.14].

3. UNCERTAINTY CALCULATION

Let us start from an analytical model that could be the chromatographic analysis of a drug which, before being submitted to the chromatographic process, has to undergo an extraction procedure. The model for the calculation of the test result is:

$$\text{drug } (\mu g/L) = \frac{(OS - a)\cdot V_{ext}}{b\cdot \text{Rec}\cdot V_{init}}\cdot f_{corr}$$

where:

OS is the output signal (for instance the area of the chromatographic peak).

a and ***b*** are, respectively, the intercept and the slope of the calibration curve (not extracted and without an internal standard).

Rec is the average recovery factor (ranging between 0 and 1).

V_{ext} and V_{init} are, respectively, the final volume after extraction and the initial volume of the sample where the measurand is dissolved.

f_{corr} is the correction factor due to the actual detector response.

The precision of a method is statistically significant if the repeated tests are independent from one another. This is not easily achieved in a single laboratory where the environmental conditions, the reagents, the instrumentation and often even

the analyst are the same. All of these are in fact factors that affect the measurement, and they do not change from one measurement to the other. For this reason, within a single laboratory the assessment must consider every single source of variability, in each step of the analytical process (bottom-up approach). On the contrary, when deriving from different laboratories, the same variability factors can be considered really independent (top-down approach). We will examine these two different approaches.

The uncertainty to be written in the final report is the "Expanded Uncertainty" which in turn is to be obtained from the "Combined standard uncertainty", calculated by means of one of the following methods.

4. PROCEDURE FOR THE BOTTOM-UP APPROACH

The uncertainty calculation must take into account all of the factors that may affect the result variability (Fig. **1**), and for each of them the factors affecting it (Fig. **2**). Figs. (**1**, **2**) are obviously only possible examples of the issues to be considered. The more the analyst's experience is, the higher the number of accounted factors will be.

The sum of all of these factors, added according to the law of propagation of uncertainty, is the combined standard uncertainty. In turn the expanded uncertainty is the product of the latter by the previously defined coverage factor *k*.

Then we may wonder: how do we assess the parameters involved in the combined standard uncertainty? First of all we must divide them into two broad categories: type A and type B contributions. In practice type A contributions are those we take into account of by the dispersion of repeated test under **repeatability conditions**, and those due to the calibration curve, which in turn is holder of its own variability. Type B contributions are linked to the intrinsic variability of the reference material, to the variability of all

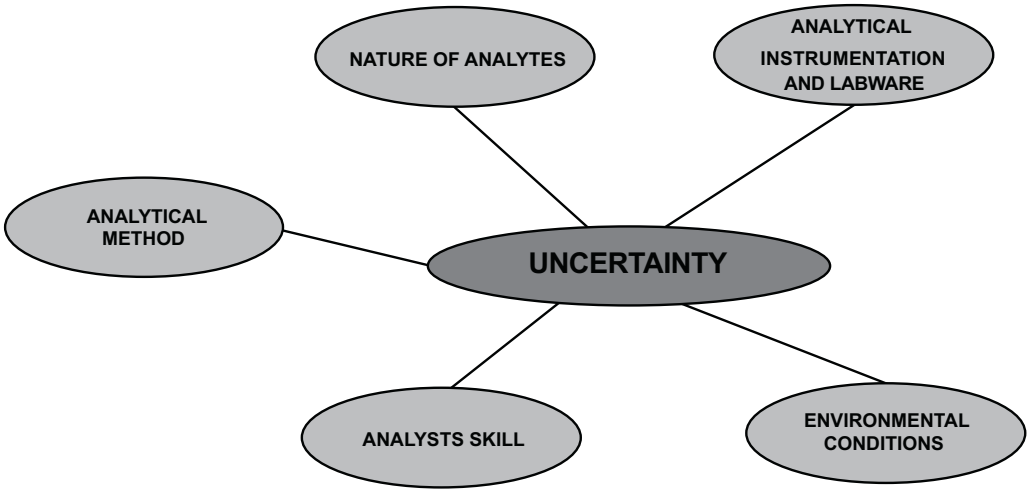

Fig. (1). Factors affecting uncertainty (general).

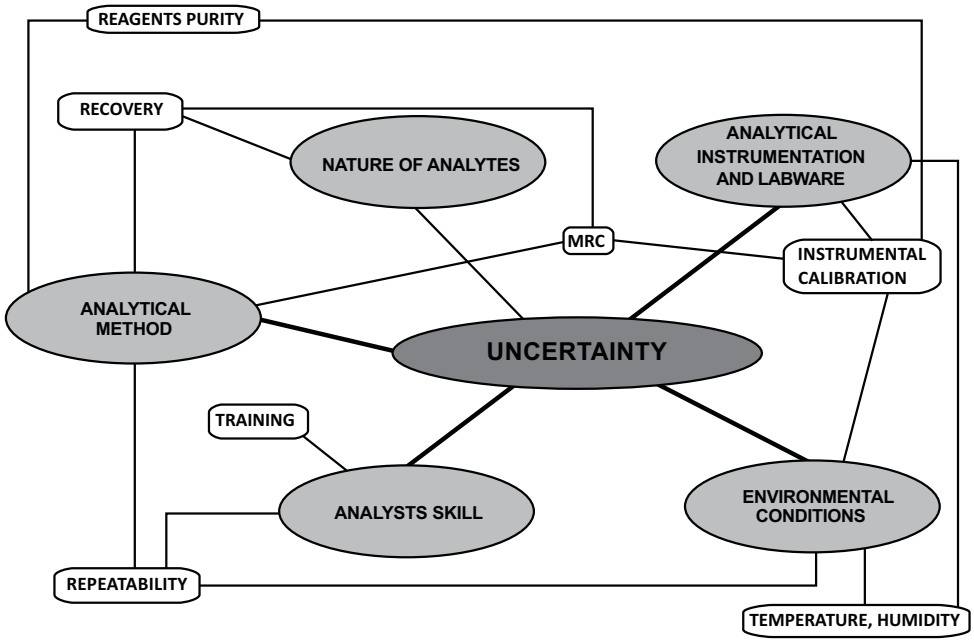

Fig. (2). Factors affecting uncertainty (closer view).

the instruments employed to measure volumes (pipettes, cylinders, flasks, etc.) or weights (for instance fluctuations of the last digit of our analytical balance), or to the changes in environmental conditions (for instance the effect of the ambient temperature changes on the glassware volumes or on the detector response).

All the above contributions are to be summed up by means of the law of propagation of uncertainty so as to obtain the combined standard uncertainty and, from it, the extended uncertainty.

5. TYPE A CONTRIBUTIONS

5.1. Uncertainty of Repeatability Calculation

First of all we have to accomplish several measurements over the same sample in repeatability conditions (at least 10). The results of such measurements are to be subjected to a test to assess the presence of anomalous data, or outliers (for instance the Dixon or Huber tests) and to another test to assess that the other data are normally distributed (for instance the Shapiro-Wilks test), usually at a confidence level of 95%. Once the unfitting data has been dumped, we have to calculate the mean $\bar{x}$ and the standard deviation s_{rep} of the random variable X of which the n survived results are as many values. At this point the uncertainty of repeatability is given by the formula:

$$u_{rep} = \sqrt{\frac{s_{rep}}{n}} \tag{1}$$

where s_{rep} is the repeatability standard deviation of the n repetitions.

The **relative uncertainty** of repeatability is thus obtained by dividing the former by the average value $\bar{x}$:

$$\dot{u}_{rep} = \frac{u_{rep}}{\bar{x}} \tag{2}$$

It is not enough yet. If the analytical routine analysis is carried out as the average of r repetitions, we ought to add the following correction:

$$\dot{u}_{rep,corr} = \dot{u}_{rep} \cdot \sqrt{\frac{n}{r}} \tag{3}$$

where n is the number of repetitions used in the repeatability calculation process (as it appears in Formula 1).

5.2. Uncertainty of Calibration Calculation

The calculation of the instrumental calibration contribution is more complex [4]. Even the points of a calibration curve are as many values – among the infinite possible – which can be obtained as the result of repeated tests over a standard of a certain concentration value, and they are all normally distributed. The calibration curve too is subdued to its own intrinsic variability (see the confidence hyperbolas theory (Fig. **3**) [5]).

Let us assume we carried out the instrumental calibration over i concentration levels, each repeated j times. Let's submit the resulting data to the linear regression function as we can find it in common spreadsheet programs, so getting the related ANOVA test value F [6]. If the test is passed, then there is a statistically significant relation between the nominal values of concentration and the signal from the instrument (for instance the count number from the mass spectrometer across the selected fragment). Let's go through another test over the intercept value to see if it is statistically equal to zero. Finally we have to apply the g function [7].

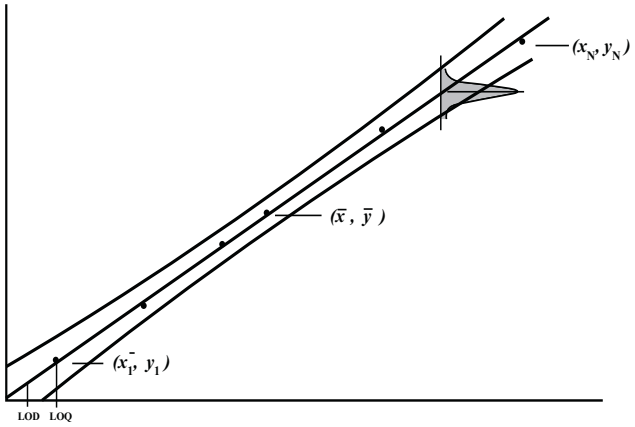

Fig. (3). Representation of a linear regression curve with the relative confidence hyperbolas.

If the test g returns a value P<0.05 then we have but to apply the following formula, with regard to one only of the n concentration values x used for the evaluation of the repeatability:

$$s_{cal,x} = s_{y/x} \cdot \sqrt{\frac{1}{r} + \frac{1}{m} + \frac{(y_{est} - \bar{y}_c)^2}{b^2 \cdot \sum_1^m (c_{ij} - \bar{c})^2}} \tag{4}$$

where:

$s_{y/x}$ is the value of the standard error or disturbance term derived by the same aforementioned spreadsheet function,

r is the number of repetitions by which the analysis is routinely carried out (see Formula 3),

m is the number of samples employed for the calibration (equal to $i \cdot j$),

b is the slope of the least square line,

y_{est} is the esteemed value obtained through the line of least squares corresponding to the concentration value x ($y_{est} = a + b \cdot x$),

$\bar{c}$ is the mean value of all the m values of concentration c_{ij} employed in the calibration curve and,

$\bar{y}_c$ is the mean value of the m experimental instrumental signals corresponding to the as many c_{ij}.

This calculation is to be repeated for each of the n samples analyzed to assess the repeatability (see Formula 1) at the correspondent level of concentration, thus obtaining n values of $s_{cal,x}$.

Once these results have been calculated, to obtain the calibration uncertainty at the concentration level x used above for the repeatability calculations, they are to be introduced into the following formula:

$$u_{cal} = \sqrt{\frac{\sum_1^i s_{cal,i}^2}{n}} \cdot \sqrt{\frac{1}{n}} \quad (5)$$

The correspondent value of the relative uncertainty of calibration is:

$$\dot{u}_{cal} = \frac{u_{cal}}{\bar{x}} \quad (6)$$

where $\bar{x}$ is the same as in Formula 2.

6. TYPE B CONTRIBUTIONS

The two aforementioned contributions are the most important ones, but other sources shouldn't be neglected either. One example is the contribution deriving from the certified reference material employed in method standardization.

6.1. Calculation of the Certified Reference Material Contribution

The formula is the following:

$$u_{CRM} = \frac{d}{\sqrt{3}} \quad (7)$$

where ***d*** is the value of the confidence interval provided by the manufacturer. For instance if the analysis certificate reported the value 1001 ± 4 mg/L then ***d*** = 4 and ***NV*** = 1001 mg/L. The relative uncertainty value is calculated as usual:

$$\dot{u} = \frac{u_{CRM}}{NV} \quad (8)$$

It can be seen that the factor $\sqrt{3}$ has been introduced because the assessment of the variability range for this parameter is not based upon statistical evaluation but on an interval characterized by two extremes out of which no value is allowed and within which every value is equally probable. We are referring to a so called "square" probability distribution (Fig. **4**, top). In other cases a "triangular distribution" is adopted, in which case the factor $\sqrt{3}$ is replaced by $\sqrt{6}$ (Fig. **4**, bottom).

6.2. Calculation of the Volumetric Labware Contribution

Let's suppose that the CRM should be diluted to obtain a working standard. This process involves volumetric hardware. For instance let's suppose that the concentrated standard is diluted 1000 times, such a dilution achieved by means of a 100 µL automatic pipette and a 100 mL glass flask.

Both of them have their nominal value and both of them have their intrinsic variability. The variability of the glass flask, for instance 100 ± 0.1 mL, is usually printed on it by the manufacturer.

The actual value and the range of the volume dispensed by the automatic pipette can be found on the calibration certificate, let's say 99 ± 2 µL. The uncertainty of the volume of the volumetric hardware affects the global uncertainty; we can calculate this by the following formula:

$$u_{flask} = \frac{0.1}{\sqrt{3}} \quad (9)$$

for the volumetric flask and

$$u_{pip} = \frac{2}{\sqrt{3}} \quad (10)$$

for the pipette. The relative uncertainties are, respectively:

$$\dot{u}_{flask} = \frac{u_{flask}}{100} \quad (11)$$

$$\dot{u}_{pip} = \frac{u_{pip}}{99} \quad (12)$$

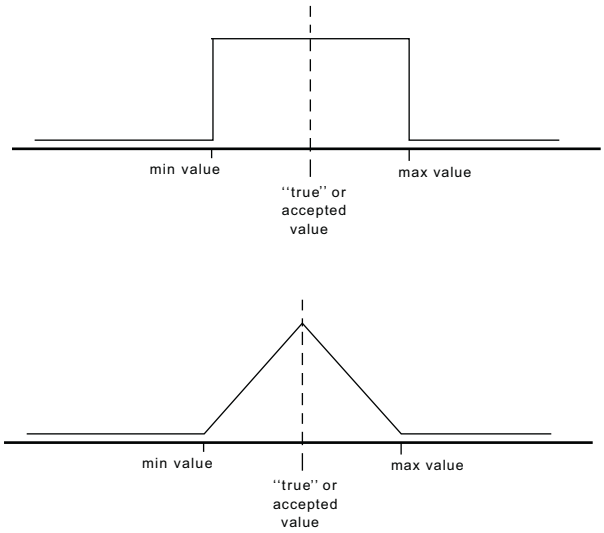

Fig. (4). Examples of "non Gaussian" statistical distributions: square distribution (top) and triangular distribution (bottom)

6.3. Calculation of Weight Contribution

If the working standard is obtained by weighing a solid CRM, the uncertainty contribution of this procedure is to be calculated. There are two types of uncertainties connected to the weighing process: the first (type A), is related to the weighing in itself: it is obtained in fact by repeated weightings at the same order of magnitude of the load we are interested in. From these measurements we get an average weight and a standard deviation that we will employ for the calculation. The second is related to the balance calibration (type B) and it is attained by the balance calibration certificate either provided by the manufacturer, or by any calibration process carried out by a specialized agency. The certificates report an extended uncertainty: the one that is of use in the sum of all the various contributions is therefore to be corrected for the coverage factor reported in the certificate, or the factor 2 if not reported. For instance if the extended uncertainty value is 0.0044 grams for a weight of 10.3 grams (tare included) the type B absolute and relative contributions of uncertainty are:

$$u_{bal} = \frac{0.0044}{2} \text{ and} \quad (13)$$

$$\dot{u}_{bal} = \frac{u_{bal}}{10.3} \quad (14)$$

6.4. Calculation of the Recovery Contribution

If the analytical method includes an extraction step we should take into account the recovery variability too (type B), at least if it is significantly different from 100%. This is if an internal standard – undergoing every step of the extraction process – is not used and we refer to a calibration curve previously achieved, that is not performed in the actual analytical session.

First of all we have to collect an adequate number of data (for instance $z \geq 20$) in intermediate repeatability conditions, whose recovery has been calculated; then we have to exclude the outliers and check if the remaining data is normally distributed. Eventually a test to assess the recovery being significantly equivalent to 100% is to be performed, through the following formula:

$$t_{calc} = \frac{|C_{CRM} - \bar{x}|}{\sqrt{\frac{s_{rep}}{n} + u_{CRM}}} \leq t_{tab:p,v} \quad (15)$$

where:

$\boldsymbol{C_{CRM}}$ is the nominal value of the CRM

is the average value of the $\boldsymbol{n}$ repetitions performed in the repeatability assessment

s^2_{rep} is the variance of the $\boldsymbol{n}$ repetitions we performed in the repeatability assessment

u^2_{CRM} is the square of the CRM uncertainty as calculated above

$\boldsymbol{t_{tab:p,v}}$ is the value of the tabulated Student's $\boldsymbol{t}$-distribution for a confidence level $\boldsymbol{p}$ and a number of degrees of freedom equal to $\boldsymbol{v}$

If the calculated t value is equal to or lower than the tabulate value, then there is no statistically significant difference between the nominal and the found value of the CRM. If this is not the case, it is necessary to take into account the recovery uncertainty, through the formula:

$$u_{\mathrm{Rec}} = \sqrt{\frac{u_{rep}}{n} + u_{CRM}} \quad (16)$$

Supposing to have considered all the contributions, we can then calculate the relative combined standard uncertainty

$$\dot{u}_{Comb} = \sqrt{\sum \dot{u}_i^2} \quad (17)$$

The absolute combined uncertainty for the selected concentration level cannot be anything but

$$u_{Comb} = \dot{u}_{Comb} \cdot \bar{x} \quad (18)$$

6.5. Calculation of the Actual Degrees of Freedom

At this point we must apply the coverage factor to attain the extended uncertainty. It is usually equal to the value of 2 when we refer to a large number of data, that is when the distribution approximates to normal. However, in the presence of fewer data it is more correct to refer to the Student's t-distribution. In this case the coverage factor depends on the number of degrees of freedom, number which is achieved by means of the Welch- Satterthwaite formula:

$$v_{act} = \frac{\dot{u}^4_{Comb}}{\sum (u_i^4 / v_i)} \quad (19)$$

where $\dot{u}_i$ are the relative uncertainties of the various contributions and $\boldsymbol{v_i}$ the related degrees of freedom. For the square distributions it is assumed that the degrees of freedom are infinite and therefore their contribution is equal to zero. The result, approximated to the nearest integer, is the one to be drawn from the tables of the Student's t-distribution for the selected confidence level. This is the sought coverage factor.

The extended uncertainty – the one to be written on the final report – is therefore:

$$U_{ext} = u_{Comb} \cdot t_{p,v_{act}} \quad (20)$$

As we can see from Formula 18 all this succession of calculations is referred to one level of concentration $\boldsymbol{x}$ only. The same procedure is to be followed for at least three concentration levels, one in the proximity of the LOQ, another near to the upper limit of linearity, and an intermediate one, if it is the case, close to a reference value, for instance the cut-off of the screening method.

Eventually it is useful to find, from the collected data, a relation between uncertainty and concentration, so as to define the uncertainty for every level of concentration within the calibration curve. If such a relation cannot be found it is necessary to divide the measurement range into a number of successive intervals within which it can be accounted for.

7. PROCEDURE FOR THE TOP-DOWN APPROACH

The top-down procedure is easier and more convenient than the former one. It is based upon the fact, already mentioned, that all the variables coming from different laboratories are completely independent. It follows that the reproducibility standard deviation is accepted as the combined standard uncertainty. This fact implies that a laboratory must take part to a collaborative trial, where more laboratories come to an agreement to test the method in question over the same sample.

Alternatively the laboratory must join a proficiency test circuit, provided that the results are returned by the organizing agency with the data from the method in question separated from all the others. That method must then be followed by every participating laboratory exactly in the same way in each and every step.

Before performing the calculation to find out the uncertainty, it is necessary to ascertain if the method performance, in the way it had been applied in our laboratory, is fitted for the intended use. To do so it is enough first to collect data apt to calculate the repeatability standard deviation $\boldsymbol{s_r}$, and then to compare it with that reported from the general data, that we will call $\boldsymbol{\sigma}$. Their ratio follows a χ^2 distribution [5]. It implies that this ratio must fall within two values, $\boldsymbol{\chi^2_{p,v}}$ and $\boldsymbol{\chi^2_{1\text{-}p,v}}$ (typically p = 0.025), as listed in statistics tables. If the resulting value is

even lower than $\chi^2_{p,v}$, then this means that the results of our laboratory are better than expected, which is good, but this fact has to be justified.

Having verified the above conditions, the reproducibility standard deviation would let us calculate the uncertainty at the correspondent concentration value. The best outcome occurs if the standard method reports a correlation function between the reproducibility standard deviation and the concentration value, or at least that it reports enough data to let us calculate such a function.

At this point the reproducibility standard deviation is simply assumed as the combined standard uncertainty of the method. The last step to obtain the expanded uncertainty is to multiply it by the coverage factor (here the degrees of freedom depend on the number of laboratories participating to the collaborative trial, and if the number is statistically significant, the coverage factor may be approximated to the value of 2).

It must be noted that in this case the uncertainty range will result, on average, larger than in the previous case because even the systematic errors of the various laboratories are randomized.

Let us see now what cases can occur in providing a result along with its range of uncertainty. All the possible alternatives are shown in Fig. (**5**) where the horizontal line represents a law limit or the cut-off of a screening method. We should consider that only in the situation as in 5a we are completely assured that the value found is under the limit, and only in the case 5g we are completely assured that we are above it. It is up to the National Scientific Societies to provide the guidelines for the correct interpretation of the results in the middle. For instance, in the case of environmental analyses, the Italian Environmental Protection Agency (ISPRA) issued a booklet [8] where they decide to consider consistent with the law limit the cases from 5a to 5d, not consistent the case 5g, and for the cases 5e and 5f they are defined "non not-consistent".

The interpretation of the numbers is up to the users or the judges. Let me give an example to better explain this concept. If we find in food a toxic substance (whose rate is regulated by law) at a level as in Fig. (**5e**), the medical authorities can decide if, for instance, it can be eaten by the adult population but, for a maximum precaution principle, should not be eaten by children.

8. CONCLUSIONS

In my opinion, the analyst's task is accomplished when they can provide a mathematically correct and statistically significant result. The interpretation is up to the users. Whatever they may adopt, they will know exactly what is the significance of the supplied number is.

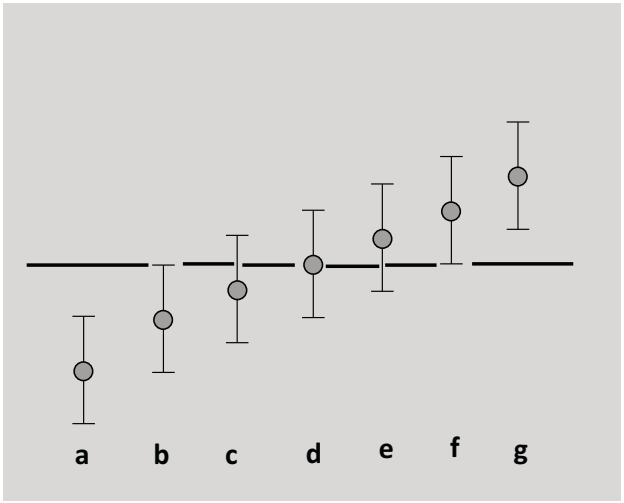

Fig. (5). Representation of different analytical results and their uncertainty in relation to a fixed reference value.

ABBREVIATION

CRM = Certified Reference Material

CONFLICT OF INTEREST

The authors confirm that this article content has no conflict of interest.

ACKNOWLEDGEMENTS

Declared none.

REFERENCES

[1] Easton VJ, McColl JH. Statistic Glossary. Sep 20 1997. Available from: http://www.stats.gla.uk/steps/glossari/confidence_interva-ls.htlm#confinterval

[2] Taylor J. An Introduction to Error Analysis. 2nd ed. Sausalito, CA: University Science Books. 1997.

[3] Bevington PR, Robinson DK. Data Reduction and Error Analysis for the Physical Sciences, 2nd ed. New York: McGraw-Hill. 1992.

[4] Tenaglia H, Venturini E, Raffaelli R. Linee guida per la validazione dei metodi analitici e per il calcolo dell'incertezza di misura. Manuali ARPA. Bologna: Industrie Grafiche Labanti & Nanni. 2002.

[5] De Martin S. Utilizzo della regressione lineare nella validazione di un metodo analitico. Boll Chim Igienisti 1999; 50: 103-8.

[6] Spiegel MR. Probabilità e statistica. Milano: Etas Libri. 1979.

[7] Miller JN. Basic statistical methods for analytical chemistry; part 2 – calibration and regression. A review. Analyst 1991; 116: 3-14.

[8] Sartori RM, Argentini D, Vannini P, *et al.* L'analisi di conformità con i valori di legge: il ruolo dell'incertezza associata a risultati di misura. Manuali ARPA. Roma: Tipolitografia CSR 2009.

Received: March 22, 2013 Revised: April 02, 2013 Accepted: April 02, 2013