DISSERTATIONES CHIMICAE UNIVERSITATIS TARTUENSIS

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ROUVIM KADIS

Evaluation of measurement uncertainty in analytical chemistry: related concepts and some points of misinterpretation

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Supervisor: Professor Ivo Leito, Ph.D.
Institute of Chemistry, University of Tartu, Estonia

Opponent: Professor Ewa Bulska, Dr. habil.
Faculty of Chemistry, University of Warsaw, Poland

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I. R. Kadis. Evaluating uncertainty in analytical measurements: the pursuit of correctness. *Accreditation and Quality Assurance*. 1998, **3**(6), 237–241.
Measurement Uncertainty in Chemical Analysis. P. De Bièvre, H. Günzler (Eds). Berlin, etc.: Springer, 2003. Pp. 147–151.

II. R. Kadis. Analytical procedure in terms of measurement (quality) assurance. *Accreditation and Quality Assurance*. 2002, **7**(7), 294–298.
Measurement Uncertainty in Chemical Analysis. P. De Bièvre and H. Günzler (Eds). Berlin, etc.: Springer, 2003. Pp. 1–7.
Validation in Chemical Measurement. P. De Bièvre and H. Günzler (Eds). Berlin, etc.: Springer, 2005. Pp. 148–152.

III. R. Kadis. Secondary pH standards and their uncertainty in the context of the problem of two pH scales. *Analytical and Bioanalytical Chemistry*. 2002, **374**(5), 817–823.

IV. R. Kadis. Evaluation of measurement uncertainty in volumetric operations: the tolerance-based approach and the actual performance-based approach. *Talanta*. 2004, **64**(1), 167–173.

V. R. Kadis. Do we really need to account for run bias when producing analytical results with stated uncertainty? Comment on ‘Treatment of bias in estimating measurement uncertainty’. *Analyst*, 2007, **132**(12), 1272–1274.

VI. R. L. Kadis. Measurement uncertainty and chemical analysis. *Journal of Analytical Chemistry*, 2008, **63**(1), 95–100.

ABBREVIATIONS

ASTM	American Society for Testing and Materials
CITAC	Co-operation on International Traceability in Analytical Chemistry
emf	electromotive force
GUM	Guide to the Expression of Uncertainty in Measurement
IEC	International Electrotechnical Commission
ISO	International Organization for Standardization
NBS	US National Bureau of Standards
RLJP	residual liquid junction potential
QUAM	Quantifying Uncertainty in Analytical Measurement
RSC	Royal Society of Chemistry
VIM	International Vocabulary of Basic and General Terms in Metrology

1. INTRODUCTION

During the last decade, the concept of measurement uncertainty has become an important new paradigm in the area of analytical quality, providing a single measure of accuracy of analytical results. In general, a measurement and, in particular, an analytical measurement cannot be properly interpreted without the knowledge of its uncertainty. Accordingly, the requirement concerning the evaluation of the uncertainty of the results produced has been included in the ISO/IEC 17025 standard [1] that establishes a quality system for analytical laboratories and also in the ISO 15189 standard [2] specifically for medical laboratories. Evaluation of the measurement uncertainty is becoming a common practice in analytical laboratories, particularly in accredited laboratories, encouraged by accreditation bodies according to the international policy [3].

Several guidance documents [4–11] from the basic ISO guide [4] and its analytical-chemical adaptation, the EURACHEM / CITAC guide [5], to the recent EUROLAB report [11] have been published, providing useful guidelines and practical examples on how the uncertainty can be estimated in a wide range of analytical applications. A few critical reviews [12, 13] mainly focus on different approaches to the evaluation process (the "bottom-up" and the "top-down") discussing their pros and cons. The problem of measurement uncertainty estimation in chemical analysis has been a central topic of several international workshops organized by EURACHEM [e.g. 14, 15] as well as seminars and courses held by other organizations around the world; and books [16, 17] covering the broad spectrum of associated problems have also been published, with the first one [16] entirely collected from journal articles.

In spite of the significant effort that went into the introduction of the measurement uncertainty methodology in analytical practice, some problems still affect its practical implementation. Analytical uncertainties are often not properly estimated, which results in a wide range of uncertainty values obtained by different laboratories for one and the same analytical procedure as evidenced from a survey [18]. In regulatory areas where analytical results are used to verify compliance / non-compliance with specified requirements, the question of credibility of the uncertainty statements has become urgent [19] as the estimates that are given often prove to be invalid. This significantly reduces the usefulness of measurement uncertainty as an index of analytical data quality. Some analysts are skeptically or even antagonistically inclined towards the applicability of the uncertainty concept in analytical chemistry [20–23].

Only partly the problems above can be ascribed to the intrinsic difficulties in the uncertainty methodology that are still under debate in analytical and metrological communities [12, 24]; rather, it is a consequence of the lack of a fundamental understanding of metrological concepts and also difficulties in practical standardization of the procedure for estimating the uncertainty. This is shown up in some points of misinterpretation in the uncertainty estimation in analytical chemistry, such as improper identification of the sources of uncertainty while considering as separate components those that do not actually make their own individual contribution to the uncertainty, or inadequate treatment of analytical bias while ignoring a persistent bias and accounting for a run bias.

Metrology in general and the issues such as measurement uncertainty and traceability, in particular, are new to most of analytical chemists, many of whom are not convinced of their value. Dr. Bernard King, the first president of EURACHEM, who made much to introduce metrological concepts in chemistry, wrote about the differences in approaches adopted in these two communities: "The gap in thinking between metrologists and analytical chemists (there are not many people who consider themselves to be both) is partly due to misunderstanding, partly to the use of differences in terminology" [25]. To this must be added that issues of quality in chemical analysis are traditionally viewed as a province of statistics, and not distinguishing between metrological and statistical ideas may lead to a significant confusion. The so called "operational definitions of uncertainty", proposed by statisticians [26], which allow the estimated uncertainty to by reduced to mere intermediate precision is an example of such confusion.

The aim of this work undertaken by the author who came to professional metrology from research in analytical chemistry is twofold. The first one is to present the notion of measurement uncertainty in the context of related concepts pertaining to metrology and quality assurance. This provides a better insight into the essence and the role of measurement uncertainty in analytical science and services. The second is to demonstrate some points of misinterpretation in the evaluation of analytical uncertainties as revealed in published guides and current literature and to provide the correct solutions of analytical problems that have not been treated properly. In a more general context, the aim the author pursued is to facilitate "bridging the cultural gap" between metrologists and analytical chemists, since a unique metrology in chemical measurement acceptable to both cultures needs to be established.

2. MEASUREMENT UNCERTAINTY AND RELATED CONCEPTS IN ANALYTICAL QUALITY ASSURANCE

2.1. The concept of measurement uncertainty in relation to accuracy and error

2.1.1. Definitions and historical overview

2.1.1.1. The definition to the term "measurement uncertainty" given in the new, 3rd, edition of the *International Vocabulary of Basic and General Terms in Metrology* (VIM) [27 (2.26)] is as follows:

> **measurement uncertainty, uncertainty of measurement, uncertainty**
> non-negative parameter characterizing the dispersion of the quantity values being attributed to a measurand, based on the information used.

This definition emphasizes the importance of the proper use of the available information about the measurand in obtaining an estimate of the uncertainty. More important is that this definition, as well as that one in the 2nd edition of the VIM [28 (3.9)], does not refer to an unknowable quantity "the true value" we are implicitly trying to find in a measurement, only focusing on the measurement result and its evaluated uncertainty. This philosophy constitutes one of the central points of the modern measurement uncertainty methodology that has being developed since the *Guide to the Expression of Uncertainty in Measurement* (GUM) [4] was appeared in 1993.

Meanwhile, the GUM itself does not exclude other, traditional, concepts of uncertainty, considering them also valid, though "as ideals" [4 (2.2.4)]. A definition of such concept can be found in the 1st edition of the VIM [29 (3.09)]:

> **uncertainty of measurement**
> an estimate characterizing the range of values within which the true value of a measurand lies.

And a still earlier wording for definition of uncertainty was given in *A Code of Practice for the Detailed Statement of Accuracy* in 1973 [30]:

> the range of values about the final result within which the true value of the measured quantity is believed to lie.

2.1.1.2. This is to say that the uncertainty is not a new concept in metrology where it was traditionally interpreted as an estimate of the likely limits to the error of a measurement. For a long time, the estimation of uncertainty was treated as an assessment of the credible error bounds around the result of a measurement. The following passage from the paper of Ch. Eisenhart [31] at the US National Bureau of Standards (NBS), dated from 1968, gives a good idea of the problem of uncertainty estimation, which has not lost its actuality in these days. “Strictly speaking, the actual *error* of a reported value, that is the magnitude and sign of its deviation from the truth, is usually unknowable. Limits to this error, however, can usually be inferred – with some risk of being incorrect – from the precision of the measurement process by which the reported value was obtained, and from reasonable limits to the possible bias of the measurement process”.

However, there was no agreement on the best method of estimation of the uncertainty components as well as the method of their final combination. Consistent with the traditional subdivision, the “random uncertainty” and the “systematic uncertainty”, each arising from corresponding sources, were propagated separately in the estimation process, and the question of how to combine them, for instance, in linear addition or quadratic addition, was an issue of debate during decades [32]. So, in a survey [33] published in 1981 seven methods for calculation of the random uncertainty (Vertrauensgrenze des zufälligen Fehlereinteils), nine methods for calculation of the systematic uncertainty (Vertrauensgrenze des nichterfaßten systematischen Fehlereinteils), and nine methods for calculation of the combined uncertainty (Vertrauensgrenze des Meßfehlers, Meßunsicherheit) were discussed and compared.

Since a standard uncertainty methodology was indispensable for engineering applications, specifically in fluid flow measurements, a compromise was then suggested and standardized by a number of organizations related to aerospace industry [34]. It was accepted to allow either linear addition or root-sum-squaring or even some other combination provided that the two components be also reported separately and the choice of the uncertainty formula be stated by the user. The books [35, 36] had summed up the progress in uncertainty methodology in engineering problems, made to the beginning of the nineties. Publication of the GUM in 1993 started a new epoch signifying the wide application of metrological concepts in various fields of science and technology.

2.1.1.3. Also not new is “uncertainty” in analytical chemistry. L. Currie wrote in 1978 [37, p. 128]: “Results of chemical analysis are of little value unless accompanied by realistic estimates of their uncertainties”. And a proper classification of uncertainty components was then put forward: „Subdivision into “inferred” and “estimated” components provides vital information on the meaning of the overall uncertainty”. It is remarkable that such subdivision was suggested irrespective of whether the error source is considered random or systematic, in much the same manner as the modern measurement uncertainty methodology treats Type A and Type B estimated uncertainties.

Of couse, the meaning of the term “uncertainty” in analytical domain changed with time. From the outset, by “uncertainty” is meant the number set down after the symbol “±” in presenting the result of analysis, which characterized the possible random error. *The Manual on Presentation of Data* [38] issued by ASTM in 1930s established the rules for calculating the "limits of uncertainty" on routine statistical basis. In the 40–50s, the assessment of these limits became the common practice in studies of analytical procedures [39, 40]. However, already to the end of the seventies, the uncertainty in chemical analysis was considered to mean a bound for the total measurement error. This meaning of uncertainty was elucidated in works of analytical chemists at NBS and expressed eventually in the definition presented by J. Taylor [41]:

> **uncertainty**
> the range of values within which the true value is estimated to lie. It is a best estimate of possible inaccuracy due to both random and systematic error.

At the same time, analytical chemists not closely related to metrology followed their own ways and used their own terminology in solving the same problem. A number of criteria for judging the acceptability of analytical results have been proposed, which combine interval estimates of random and systematic error one way or another. Mention should be made of „total error" [42], „total analytical error" [43], „maximum total error" [44], and others (see Ref. 44).

2.1.2. Uncertainty and accuracy. Difference in philosophy: a chemist's view

The last definition above says that uncertainty is simply an estimate of possible "inaccuracy". The positive connotation of this term – "accuracy" – is in common use by analytical chemists for many years. Accuracy has always been considered the most important concept characterizing the quality of analytical methods and results, recognized as a capital analytical property in the hierarchy [45]. The question of interrelation between two concepts, accuracy and uncertainty, is therefore worthy of consideration.

2.1.2.1 According to its definition in the VIM [27 (2.13)]:

> **measurement accuracy, accuracy of measurement, accuracy**
> closeness of agreement between a measured quantity value and a true quantity value of a measurand,

accuracy is a qualitative concept. Yet, it can be quantified if described in terms of "precision" and "trueness" corresponding to random and systematic errors, respectively. (A survey of the evolution of these terms and concepts in standardized terminologies is presented in [46].) Following this line, the two measures of accuracy, the estimated standard deviation and the (bounds for) bias, are to be evaluated and reported. As the traditional theory of measurement errors holds [47], these two figures cannot be rigorously combined in any way to give an overall index of (in)accuracy in view of the different nature of the error components. And it is in this way – as a two-component vector – that the (in)accuracy of analytical methods and results was traditionally evaluated in analytical chemistry [37, 48–50]. This methodology has been embodied in the IUPAC harmonized protocol for method-performance studies [51] and the ISO 5725 series of International Standards [52].

2.1.2.2. In a recent paper [53] on the rationale for the 3rd edition of the VIM, two different approaches to the philosophy and description of measurement have been formulated, called there the “classical approach” and the "uncertainty approach”.

The classical or traditional approach, as it was discussed as far back as 1971 [54], is based on two postulates:

α – there exists a true value of the quantity measured;
β – the true value is impossible to seek.

The first statement appears as a necessary prerequisite and the objective of a measurement: starting a measurement, we proceed from the belief that some value of a measured quantity exists, which we are trying to find. The second statement says that our effort will result in only an approximation, i.e. an estimate of the true value. This is equivalent to the fact that measurement errors are inevitable.

The uncertainty approach in its turn, with the GUM as its most prominent representative, insists that since it is not possible to know the true value and, furthermore, there may be a set of true values that are consistent with the definition of the measurand, it is not possible to know how close a value obtained is to the true value. Thus, “measurement error” becomes an abstract concept, useless in practice. What is more important as a practical matter, this approach provides a quantitative means of combining information arising from both random and systematic “effects” (the word “error” is not used here) on equal footing, treating all of them as standard-deviation-like quantities after the corrections for known systematic effects have been made. As a result, a variance of a probability distribution function (PDF) is estimated that is used to characterize the quality of a measurement, based on all the information used.

2.1.2.3. It seems that the fact that the uncertainty approach implies a radically different standpoint in philosophical foundation of measurement has escaped the attention of researchers. This approach actually means (see Ref. 55) a transition in concepts from the statement “numbers are in the world” (J. Kepler), and indeed “in measurement, numbers are discovered rather than assigned” [56] to the statement that “numbers are assigned to the nature by ourselves” (R. Carnap), so that measurement is not a determination but an *assignment* (of numbers). This is a radical change of the general conceptual basis of measurement, with the very concept of true value disappearing as simply useless.

There has been a long-standing debate about the reality of true values: do "true values" really exist if they are empirically inaccessible? Not long ago the question was raised with regard to analytical measuremnt [57]: "Is there a "true value" in nature or in our experiments at all?" And if not, shouldn't we forget about "true values"?

Analytical chemists who actually deal with *determination* (of the chemical composition of a material) are unlikely to be ready to take these challenging ideas. When speaking about "true value" the chemist has in mind the actual correct composition. And since chemical entities (molecules, ions) are discrete entities that can in principle be counted, the correct composition certainly exists. Counting molecules is becoming increasingly possible also in practice (e.g. on a surface, using an atomic force microscope) making the true materials composition accessible. In many cases the analyst can set up the true value himself by adding a known amount of the pure substance to the blank sample; thus the reality of the true value is beyond question for chemical way of thinking.

2.1.2.4. Prof. Paul De Bièvre, one of the most consistent proponents of the new philosophy in analytical measurement, argue [58] that with the evaluation of uncertainty, "a doubtful meaning of 'accuracy' (doubtful because tied to 'true value') is replaced by a practical one: 'measurement uncertainty'", with no longer any necessity for mentioning "accuracy" and "true value" as unproductive concepts. However, as discussed in [53], what is actually done in the uncertainty approach is that an interval is established "within which the 'essential unique' (true) value of the measurand is thought to lie, with a given probability…". The concept of true value is indeed necessary for formulating the objective of measurement, but it is not directly used in the evaluation of the uncertainty interval. The question is whether it is justified to banish "true value" from our consciousness for the reason that we do not use it in the calculations.

In my opinion, discarding the traditional concept of accuracy with regard to uncertainty evaluation is neither helpful nor necessary. It is seems quite appropriate to use the philosophy of the classical approach combined with the tools and practical methodology provided by the uncertainty approach. In this way, the measurement uncertainty can be viewed as a single-number index of accuracy, being constructed following definite rules.

This viewpoint on the relationship between "accuracy" and "uncertainty", put forward in the original publication II, has found corroboration by the RSC Analytical Methods Committee who recognized that "an estimate of uncertainty is probably the most appropriate means of expressing the *accuracy* of results" [59].

2.1.3. Random, systematic, and “systematic” error

The subdivision of measurement error into random and systematic is fundamental for the traditional approach where the accuracy is estimated in terms of precision and trueness. In contrast, following the GUM, all the uncertainty components are combined as random quantities, using their squared standard deviations. “The denial of the principal difference between random and systematic errors” furnished the main point of objection raised by the opponents of the application of the uncertainty methodology in chemical analysis (see e.g. Ref. 22 from which the quotation was taken). The detailed discussion that follows, based on the original publications II and VI, is concerned with this topical issue.

2.1.3.1. To a large extent, the classification of measurement errors with any of these types is a matter of convention; it depends on the level from which the analytical system is being viewed. An error that appears to be systematic at one level becomes random at a higher level of the system. For example, laboratory bias (in performing analyses by a certain procedure) becomes a random error if we change to the consideration of a multitude of laboratories. V. Nalimov wrote in [60] in this connection: “It is possible to speak about random errors only when the set of measurements is clearly defined and limited.”

A good example of such a limitation is found in the definition of “random error” given in the VIM, 2nd edition [28 (3.13)], which relates to measurements made under repeatability conditions (the same operator, the same equipment, a short period of time), i.e., the conditions within an analytical run. With this strategy, the unambiguous categorization of errors is made possible. Measurement error characteristics estimated under other experimental conditions are to a lesser or greater extent “contaminated” by systematic error components.

2.1.3.2. The conditional character of error subdivision into random and systematic can be seen from the hierarchical model of analytical errors proposed by M. Thompson [61]. According to this model, the error in the result of analysis of a homogeneous material consists of the following components: (1) the method bias, (2) the laboratory bias, (3) the run bias, and, finally, (4) the random error (under repeatability conditions). This model referred to as a “ladder of errors” is considered as the basis for assessing the uncertainty of the results, which is obtained by combining the uncertainty contributions of the steps of the ladder. It is significant that all of the error components listed above, and those which appear as “bias”, are treated [61] as random variables characterized by their standard deviations.

2.1.3.3. The subdivision into random and systematic errors results from the “technology” of experimental estimation of accuracy, when a material of a known composition is analyzed according to a given procedure. The indexes of precision and trueness so obtained characterize the *quality of the analytical procedure*. However, the customers, i.e. those who use the results of analyses for any purpose, are interested in the *quality of the results*, expressed in a number. From this point of view, it does not matter if the error of the result is random, systematic, or contains both constituents. Focusing on the quality of the product, i.e. the result of analysis, puts a characteristic of the total error at the forefront.

2.1.3.4. By systematic error is traditionally meant "the measurement error component that remains unchanged or changes regularly upon the repeated measurements of one and the same quantity" [62]. However, because of the impossibility of the precise reproduction of a whole set of experimental conditions, scientists have long come to realize that the systematic error usually includes both the constant and the variable component. As discussed by Ch. Eisenhart [47], even in the case of repeated measurement of a single distance, some effects, appeared as systematic, will be constant, but some may be expected to vary from one occasion to the next (e.g., from day to day), thus contributing *variable components* to the *systematic error*. In chemical analysis, these error components have long been known as "veränderliche (schwankende) systematische Fehler" [63], "variable part of systematic error component" [64], or even "random components of systematic error" [65]. An important point is that the constant (strong systematic) error component, when it is estimated and known, can be eliminated by applying a correction to the result. But the variable component, as a random quantity, should be accounted for along with the common random error contribution.

2.1.3.5. These ideas were developed in the conception of randomization of systematic errors [66, 67] just in the context of the problem of estimating the measurement uncertainty. Accordingly, what was traditionally referred to as systematic error is treated as one special value taken at random out of a population of all possible values of the systematic error for the experiment under consideration. In this way, a peculiar random variable, so-called "systematic" error, is introduced with a respective probability distribution assumed. Quantifying the variance of such a (hypothetical) distribution, when there is no statistical information, is a special issue. In this case, one has to use subjective estimates based on experience and available knowledge. But this approach makes it possible to treat all error contributions, both random and "systematic", on equal footing and combine them as corresponding variances into the variance of the resulting probability distribution.

2.1.3.6. Let us assume that the point of interest is the accuracy of the result of analysis that has been (or will be) obtained using a certain procedure; it is not necessary that this was (or will be) done on the given instrument, by the given analyst, or even at the given laboratory. In other words, we are interested in the accuracy of the results obtained using a multitude of possible implementations of the procedure rather than one implementation. In such a formulation of the problem, the local systematic errors inherent to each of those implementations appear as values of a random variable, "systematic" error, not fundamentally different from the random error in the strict sense. The distinction is only that it is defined on a population not identical with that of the observed measured values.

This situation is typical of so-called technical measurements (commonly performed in control engineering by technicians) when the results are obtained following specified procedures, with the accuracy evaluated before the measurement is made. Classification of errors as random and "systematic", adopted in technical measurements [68], provides a unified method for combining the constituents (as squared standard deviations) in calculation of the total uncertainty. The same principles are applicable in analytical measurements where an analyte is determined by following a specified procedure stated, for instance, in a written standard. It is such measurements that are usually referred to as routine in chemical analysis.

2.1.3.7. We can state with confidence that the concept of "systematic" error introduced in physical measurement is particularly appropriate in chemical analysis. This error occurs here on a set of implementations of two types, a set of procedure implementations and a set of samples to be analyzed by the procedure and differing in their matrix composition or physical properties, which affect the measurement result. Indeed, the difference between the test samples and the reference material in these properties results in the appearance of a "systematic" error usually associated with the inadequacy of calibration; this is typical for chemical analysis.

2.1.3.8. In analytical measurement, the matrix mismatch is always attendant when one analyses an unknown sample "with the same matrix" using a fixed, previously determined, calibration function. Usually, an analytical procedure is developed to cover a range of sample matrices so that an "overall" calibration function can be used. An error due to matrix mismatch is therefore inevitable if not necessary significant. It constitutes in fact an inherent part of the total variability associated with the analytical procedure.

Meanwhile, these effects are in no way included in the usual measures of accuracy as they result from a "method-performance study" in accordance with the accepted protocols [51, 52]. The accuracy experiment there defined (Ref. 52, Part 1, Section 4.2) does not presuppose any variable matrix-dependent contribution, being confined to identical test items. The underlying statistical model assumes that solely laboratory components of bias are to be considered.

Error sources of this kind are just the ones fairly treated with the concept of measurement uncertainty which makes no difference between "random" and "systematic". By using simulated samples the effect of the matrix is a matter of direct investigation in respect of factors that influence the result and may be at different levels for analytical samples and calibration standards. The experimental designs for such studies, which provide for variation of matrix factors at least at two levels corresponding to their upper and lower limits, have long since been suggested [69, 70].

2.1.3.9. The conclusion to be made is that traditional subdivision of errors into random and systematic is inconsistent with the present view of the error structure in measurements that follow specified procedures where the peculiar random variable, “systematic” error, occurs. This is typical of analytical measurements especially.

It would be more appropriate to classify the errors as *corrigible* (those about which we may do something) and *incorrigible* (those about which we can do nothing). Both of the random and “systematic” components would be merged together in the second group, then. Such classification of errors, which was suggested long ago [71], is in agreement with the GUM principles of uncertainty evaluation. Indeed, an estimate of the corrigible error is used to make a correction; the incorrigible error contributes to the uncertainty of the result, with the uncertainty of the correction taken into account as well. This is illustrated schematically in the following diagram (Fig. 1).

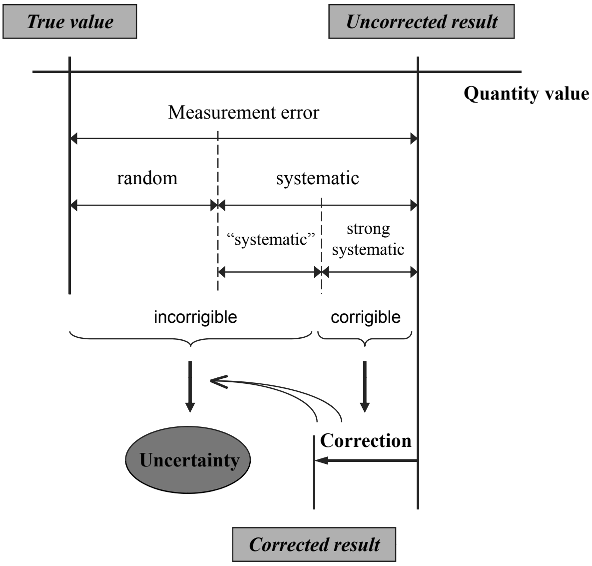


Figure 1. Classification of measurement errors and evaluation of uncertainty

2.2. Analytical procedure – the key concept of quality assurance in chemical analysis

2.2.1. Measurement assurance, measurement uncertainty, and measurement procedure

K. Doerfell referred to analytical science as a discipline between chemistry and metrology [72]. In line with this, one may define analytical services – a sort of analytical industry performing chemical analyses for customer needs – as a system based upon concepts of chemistry, metrology, and industrial quality control.

2.2.1.1. It is of principal importance in analytical work in service to view measurement from a perspective of a "production process", so that the measurement result be regarded as the output of a repeatable process, analogous to an industrial production process, also designated the chemical measurement process. Then we can apply the principles and techniques of quality assurance developed in physical measurement to analytical work. These principles and techniques constitute the field of *measurement assurance* [73], a system affording a confidence that the measurements produced in a measurement process are valid, that is, good enough for their intended use.

From metrological standpoint, "good enough" means simply having an allowable uncertainty. If measurement serves any practical need, it is to meet some requirements on accuracy / uncertainty, emerging from the purpose. This is a quotation from the NBS report [74] dated from 1974: "For each practical measurement it is only necessary that the uncertainty of the result be adequate for this intended usage". It is in this case that the result of a measurement is considered "fit for purpose".

2.2.1.2. In effect, the uncertainty associated with a sequence of operations defined as a measurement is to be determined; this sequence of operations by which the measurement is made constitutes the *measurement procedure*.

The characteristic feature of the measurement uncertainty methodology is that any numerical measurement is not thought of in isolation, but in relation to the process which generates the measurements. If all the factors operative in the process are defined, they actually determine the relevant uncertainty sources, so making practicable their quantification to finally derive the value of the total uncertainty. It can be said that the uncertainty methodology as presented in the GUM fits neatly the idea of a specified measurement procedure, because the procedure defines the context the uncertainty statement refers to.

Two basic propositions underlie this strategy in chemical analysis. First, an analytical procedure when performed as prescribed, with the chemical measurement process operating in a state of control, has an inherent accuracy to be evaluated. Second, a measure of the accuracy (in this case – measurement uncertainty) typical of a given procedure can be transferred to the results produced, providing a degree of their confidence. The justification for both of the propositions was given by W. Youden in his works on analytical performance [75, 76].

2.2.1.3. As a prerequisite for practical implementation of this approach it is assumed that the chemical measurement process remains in a state of statistical control, being operated within the specifications. “Control” means that performance characteristics of the process, namely, precision and bias, remain fixed although they cannot be determined exactly.

To furnish evidence for this and avoid reporting invalid data the analytical system needs to be tested for continuing performance. A number of control tests may be used with this aim, for instance, testing the difference between parallel determinations, duplicating complete analysis of a current test material, and analysis of a reference material.

An important point is that the control tests be performed in such a manner and in such a proportion as a given measurement requires, comprising an integral part of the whole analytical procedure. They will be more specific in this case and relate to critical points of the measurement process.

2.2.1.4. It is significant in this scheme that measurement uncertainty is estimated during procedure development and validation, before its regular use, and the uncertainty is assigned to future result obtained by application of the procedure under specified conditions. This approach is common to measurements referred to as routine in chemical analysis as discussed in paragraph 2.1.3.6.

2.2.2. Analytical procedure in terms of quality assurance

One of the ways to facilitate an effective quality assurance strategy in analytical chemistry is integrating quality aspects into common analytical concepts. The notion “analytical procedure” seems to be that best suited to such a quality-oriented (re)formulation.

2.2.2.1. The proper usage of the term “procedure” rather than “method” is noteworthy first. These terms correspond to two adjacent levels of specificity in the hierarchy of analytical methodology [77, 78] expressed as a sequence from the general to the specific:

technique ⇨ method ⇨ procedure ⇨ protocol

According to this, the *procedure* level provides the specific directions necessary to utilize a *method*.

This nomenclature is however not always adhered to. In many cases, an analytical procedure is virtually implied when an analytical method is spoken about. Written standards are usually titled “method of analysis”, although they provide directions described specifically to be followed in their application. Such expressions as “validation of analytical methods” or “performance characteristics of analytical methods” are typical examples of incorrect usage of the terms.

On the other hand, there is normally no reason to differentiate two of the most specific levels in the hierarchy, carrying the term “procedure” over to the designated “protocol”. According to the definitions of the two notions in [77], specific written directions have to be faithfully followed in both cases. The term "standard operating procedure" (with emphasis on the standardized format of the document) that is used in quality assurance terminology and also mentioned in VIM as a synonym of “measurement procedure” [27 (2.6)], seems to cover the two levels in the hierarchy.

2.2.2.2. A distinction needs to be drawn between analytical procedure as a general concept and its particular implementation, i.e. an individual version of the procedure embodied under particular circumstances. In practice, an analytical procedure may exist as a variety of implementations, differing in terms of equipment, reagents, environmental conditions, and even the analyst’s own routine if all of them are within the stated specification. As explained in paragraph 2.1.3.6, this variability is a characteristic source of uncertainty associated with a measurement procedure.

2.2.2.3. In the ordinary sense the term “analytical procedure” means simply a description of what has to be done while performing an analysis. The definition to this notion referenced in [79] is rather diffuse: “The analytical procedure refers to the way of performing the analysis. It should describe in detail the steps necessary to perform each analytical test. This may include but is not limited to: the sample, the reference standard and the reagents preparations, use of the apparatus, generation of the calibration curve, use of the formulae for the calculation, etc.” [80]. In brief, this means a description of everything that should be done in order to perform the analysis.

The reasoning in paragraph 2.2.1.2 gives grounds to include a quality assurance aspect in the notion. The case in point is not simply “the way of performing the analysis” but that one which ensures obtaining the results of a specified quality. An accuracy/uncertainty constraint should be built in the definition so as to give a determining aspect of the notion. Thus we come to the definition given in terms of quality assurance: a *specified procedure to follow which ensures obtaining the results of analysis with a known uncertainty.* This is schematically depicted in Fig. 2 where typical “constituents” of an analytical procedure are shown.

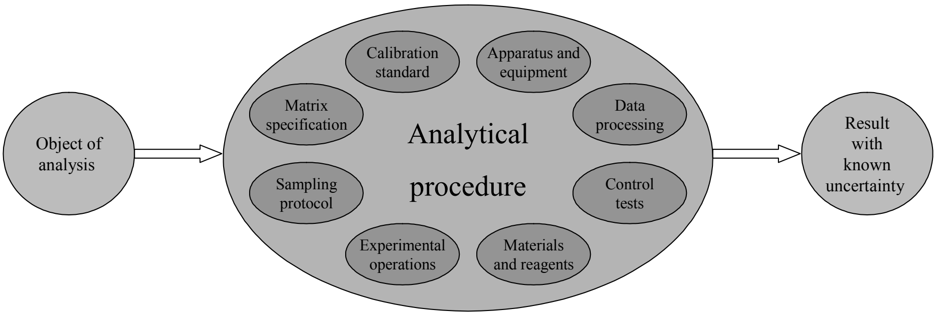


Figure 2. Typical "constituents" of an analytical procedure, to be specified, which ensures obtaining the results with a known uncertainty

2.2.2.4. The concept of analytical procedure so defined offers an advanced view on the problem of "method validation". By this term is generally meant a process of establishing the performance characteristics and limitations of a method and verifying that the "method is fit for purpose" [81], i.e. suitable for solving a particular analytical problem. The question is how the suitability should be assessed, based on customer needs.

It is commonly recommended [e.g. 80, 81] that a number of characteristics such as precision and trueness, limits of detection and quantitation, selectivity, sensitivity, and so on, be considered as criteria for analytical performance and evaluated in the course of a validation study. Based on this, a judgement is made as to whether the procedure under issue is capable of meeting the specified analytical requirements. However, from the customer point of view the matter of primary concern is quality of analytical results as an end-product. In this respect, a procedure will be deemed suitable if the data produced are only fit for purpose.

It follows that measurement uncertainty is the parameter that needs paramount consideration among others in validation studies. Other characteristics are desirable to ensure that a methodology is well-established and fully understood, but validation itself on those criteria seems impractical also in view of the lack of corresponding requirements as is commonly the case. That is to say that the evaluation of uncertainty and the assessment of compliance with the uncertainty requirements shall constitute the ultimate goal of validation. With analytical procedure as defined above a conceptual framework for such validation process is being developed.

This standpoint originally presented in the publication II has found corroboration in the IUPAC guidelines [82] on single-laboratory validation, which states that "…method validation is tantamount to the task of estimating uncertainty of measurement". The recent proposal [83] to replace the traditional set of performance characteristics, on which an analytical procedure is selected, with the single "uncertainty function" that describes how the uncertainty of measurement varies with the analyte concentration represents the motion in the same direction.

2.3. The GUM and the “whole method” performance methodology in uncertainty estimation

2.3.1. The modelling approach and the empirical approach

2.3.1.1. The GUM principles form a consistent and widely applicable methodology where the uncertainty is calculated by a combination of individual contributions from different sources of uncertainty. This propagation procedure implies the identification of all relevant uncertainty sources, the estimation of their magnitude, and combination of the components to form the combined standard uncertainty, and then, the expanded uncertainty. Concurrently with this, possible sources of bias (the error depicted as strong systematic in Fig. 1) are investigated, and corrections for significant systematic effects are assumed. So, a detailed analysis of the factors influencing the result and a quantitative model in which those factors appear as input quantities are required.

2.3.1.2. In spite of the helpful adaptation of the GUM specifically for analytical chemical problems, made in the guide *Quantifying Uncertainty in Analytical Measurement* (QUAM) [5], and the extension of the GUM principles to the field of quantitative testing [7, 10], this methodology received approval mainly among “metrology-oriented” analytical chemists. Indeed, this approach differs significantly from that traditionally being used for assessing the “whole method” performance. The performance characteristics such as precision and trueness are obtained during the procedure development and validation within a laboratory or in a collaborative study. In that case neither analysis of influence factors nor mathematical model is required, since the approach is entirely empirical.

2.3.1.3. This corresponds to the role of models in two principal approaches to measurement, classical and uncertainty methodologies, discussed in 2.1.2.2. In general, the uncertainty approach emphasizes the model-dependence of the knowledge obtained by measurement, while the classical approach tends to diminish the role of models. As regards analytical measurements, we can say that an exhaustive model of a chemical measurement process is not always possible to build because of the complexity of the process. It is not surprising that an analytical procedure is treated as a black box, i.e. a system with unknown internal structure, in the system theory [84]. Turning a black box into a white (transparent) box is not so easy, which is why the “whole-method” performance studies have been traditionally used in analytical chemistry.

2.3.1.4. It was suggested and substantiated by the RSC Analytical Methods Committee [85] to consider the traditional “whole method” performance studies as a “top-down” method of dealing with uncertainty in contrast to the GUM-based “bottom-up” method. With the top-down approach, a laboratory is viewed from a “higher level”, i.e. as a member of population of laboratories, and the collaborative study is assumed to take a representative sample of combinations of operating influence factors. It follows that the reproducibility standard deviation obtained in the collaborative study is a good estimate of the combined uncertainty that would be obtained following the GUM. In fact, a direct collective quantification is used instead of individual quantification and combination of uncertainty components.

2.3.1.5. Most recently, another classification scheme was presented by EUROLAB [11] in which the GUM-based methodology is referred to as *modelling approach* in contrast to *empirical approach* that encompasses three “whole method” performance methodologies: single-laboratory validation, interlaboratory studies, and proficiency testing. There is a clear tendency toward involving a variety of quality assurance means in the uncertainty estimation process, which meets the actual demands of laboratories and more adequately reflects common analytical practice.

2.3.1.6. The modelling approach based on an exhaustive analysis of the measurement system, modelling, and propagating uncertainties is metrologically sound, and the outcome of only this evaluation process can be called “measurement uncertainty” in the strict sense. On the other hand, the empirical approach makes allowance for the all-inclusive variability at once, but immediately poses the questions of sufficiency, the extent of coverage, and “extrapolation” from the specific conditions of the performance study to the actual conditions. There are sources of uncertainty such as approximations/ assumptions incorporated in the measurement method which are fundamentally difficult to be allowed for empirically if staying within the given method.

2.3.1.7. The EUROLAB report [11] asserts that the empirical approaches to measurement uncertainty are as valid as the modelling approach. It is stressed however [11 (1.1.3)] that with any approach 1) the measured quantity should be clearly and unambiguously defined, and 2) the measurement process should be carefully analysed “in order to identify the major components of uncertainty”. In other words, a list of relevant uncertainty sources is required to be given. Thus, the first two steps in the GUM methodology, namely, specification of the measurand and identification of uncertainty sources, are transferred to and become necessary in the empirical investigation. Only in this way, the compliance with the GUM principles, which is declared in [11], may be achieved.

2.3.2. Rational strategy

2.3.2.1. In view of complexity of the problem, the GUM methodology in its pure form is difficult to implement in analytical measurement. If a combined effect of several factors is observable, the relevant estimates of the overall variability and bias can reasonably be taken into account in the uncertainty budget. The worked examples presented in the second edition of the QUAM [5] provide a good illustration of such kind of mixed approach.

It is indeed an effective expedient in analyzing different uncertainty sources not to isolate random variations intrinsic to individual sources, but instead grasp them as an experimental repeatability estimated on replication of the procedure as a whole. The same is true with regard to bias and its associated component of uncertainty, usually studied experimentally for the whole procedure. The appearance itself of the entries "repeatability" and "bias" (or "recovery") in uncertainty budgets and of the corresponding branches in the cause and effect diagrams in [5] signifies a shift to the empirical approach from the pure GUM-based methodology. Can we say that we strictly follow the GUM in identifying uncertainty sources as *input* quantities in the measurement model, if the precision characterizing the *output* quantity is directly estimated and displayed among the sources of uncertainty?

2.3.2.2. The rational strategy to follow is in using different approaches conjointly, that is, combining the available "whole method" performance data with additional uncertainty contributions not adequately covered in the experimental design. It is appropriate to use in this combination process the "highest level" precision measure available, the reproducibility standard deviation from a collaborative trial, which would require a minimum amplification by additional uncertainty contributions. Often, however, one has to use merely the repeatability estimate, to which other required components are added. In any case the use of performance information, at least the repeatability standard deviation, is essential.

2.3.2.3. Some issues need to be addressed in using data from prior studies, such as relevance of the data, variation in conditions, pre-treatment steps which may not have been included in the study, and others [86]. It is recommended [87, 88] that all these questions be answered in a reconciliation stage [5 (7.2.1)] where the available information is compared with the required one (which is based on the analysis of uncertainty sources). Any factors that affect the measurement but have not been taken into account during the empirical investigation should additionally be assessed for their contributions to the overall uncertainty.

2.3.2.4. Thus, we have to deal with a combination of different methodologies in uncertainty estimation. The recent guides issued by ISO [8], NORDTEST [9], and EUROLAB [11], which follow this general strategy, demonstrate such an integrated approach to estimating measurement uncertainty in chemical analysis.

3. SOME POINTS OF MISINTERPRETATION IN THE EVALUATION OF ANALYTICAL UNCERTAINTIES

3.1. Quantification of uncertainty and correction factor in a multistep analytical procedure

3.1.1. The trueness of an analytical procedure is often expressed in terms of recovery, i.e. the ratio of the measured value of the quantity to the expected (reference) value. This is justified where the relative measurement errors are approximately constant over the entire measurement range. In this case the analytical bias may be corrected using a correction factor by which the result is multiplied.

3.1.2. In principle, for an analytical procedure consisting of *n* successive steps, it is possible to derive the combined uncertainty u_C and the combined correction factor F_C from the component uncertainties u_i and correction factors F_i, each relating to a step *i* as follows:

$$u_C = \sqrt{\sum_{i=1}^{n} u_i^2} \tag{1}$$

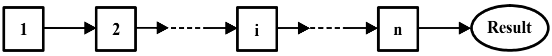
$$F_C = \prod_{i=1}^{n} F_i \, . \tag{2}$$

This method was used specifically in Example A3 “Determination of organophosphorus pesticides in bread” in the first edition of the guide QUAM [89]. The values u_i and F_i for different steps in the procedure, such as liquid extraction, cleaning-up and concentration of the extract, and so on, were obtained there from recovery experiments: if R_i is the recovery on the step *i*, the correction factor $F_i = 1/R_i$.

3.1.3. The formulas (1) and (2) are only true if the components combined represent individual contributions which are strongly independent. This is not the case in that example, however. For example, the recovery R_3 at the extraction step is determined by carrying out several operations, beginning with extraction and ending with gas chromatography determination. The correction factor F_3 will consequently include systematic effects on the steps that follow the extraction. Likewise the uncertainty u_3 incorporates all the succeeding uncertainties. In this way, *summary* estimates are actually used in the calculation instead of *individual* ones, which makes the estimation of the combined parameters wrong.

3.1.4. To get the individual correction factor F_i for a step *i* one must divide the available summary estimate $\widetilde{F}_i$ by the correction factor(s) relating to all the subsequent steps and then, one can calculate the combined factor F_C for the whole procedure. Alternatively, it is possible to do this without finding the individual estimates for all *n* steps if to use as a factor in the product the summary estimate $\widetilde{F}_i$ covering the steps from *i* to *n*.

The diagram on Fig. 3 demonstrates the two possible ways in the calculation of the combined correction factor F_C and the combined uncertainty u_C. Deriving individual estimates from summary estimates is depicted by upright arrows, and obtaining the combined parameters as a product or root of sum of squares is depicted by horizontal arrows in the diagram. Figures relating to a step *i* (shown in circles) are either individual estimates F_i, u_i (upper part of the table) or summary estimates $\widetilde{F}_i$, $\widetilde{u}_i$ (lower part of the table).



Scheme of a multistep procedure

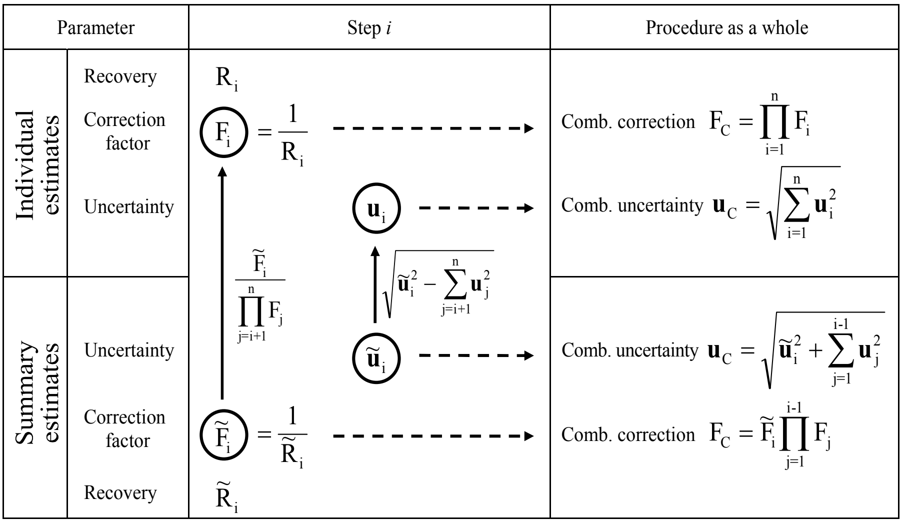


Figure 3. Evaluation of combined correction factor and combined uncertainty in a multistep analytical procedure

3.1.5. This pattern demonstrates that the process of propagation of uncertainties needs great care to be correct. To avoid complications, we have to identify the components as resulting from individual (or combined) *sources of uncertainty* rather that from separate *parts of the analytical system*, which are difficult to be evaluated in isolation. The case of a multistep analytical procedure where the effects of individual steps are evaluated jointly from the final measurement result illustrates this feature clearly.

The criticism on the evaluation of the uncertainty und the correction factor in Example A3 in the guide [89] was drawn in the author's publication I. In the second edition of the guide [5], this example (now as Example A4) has been totally revised with emphasis on the analysis of possible sources of uncertainty and their quantification utilising the data from in-house validation studies.

3.2. Volumetric uncertainty: the tolerance-based approach and the actual performance-based approach

3.2.1. Being a part of most analytical procedures, measurement of volume belongs to basic analytical operations. This is commonly performed by using volumetric glassware that is the second most fundamental type of measuring instruments in an analytical laboratory, after the balance. The question of the accuracy of volumetric operations has always been topical and has always been answered in terms of the specified tolerances to which the glassware is manufactured (see e.g. Table 4.1 in [90]).

3.2.2. The EURACHEM guide [5, 89] (in worked examples, Appendix A there) has stated that three separate contributions should be taken into account when volumetric uncertainty is estimated:
1. Capacity tolerances for volumetric apparatus, taken from the specification.
2. Random variation from a volumetric repeatability experiment.
3. Ambient temperature effects.

At this point, two questions arise. The first one concerns the random variation inherent in the use of volumetric apparatus: has it not really been included in the stated tolerance? The second reads as follows: if nevertheless the actual performance has been studied, why is it used in calculating the uncertainty along with the tolerance, not instead of it, resulting in redundancy in uncertainty estimation?

3.2.3. Volumetric apparatus that are produced commercially shall meet certain metrological requirements, normally in the form of "maximum permissible error", specified by a written standard to which the apparatus is made. If the compliance with the specification is originally guaranteed by the manufacturer or demonstrated by an independent authority, it is accepted that the errors produced *while the instrument is in use* do not exceed its specified limit of permissible error. This is a basic tenet of the quality assurance system established for measuring instruments in various measurement fields, including volume measurement.

3.2.4. The use of volumetric apparatus involves a series of operations such as filling the vessel, setting or reading the meniscus against a reference line or scale, and draining if the device is intended for delivery. In other words, any volumetric measurement is associated with some operating procedure that an analyst has to implement. This leads to the situation where the accuracy inherent in volumetric apparatus cannot be evaluated in isolation, by ignoring the contribution of a procedural error that is inseparable.

If all significant error sources are kept under control by following a standard procedure, the procedural error may be assumed to lie within a limit. It is therefore conventional for volumetric glassware to incorporate the random error contribution, typical for its proper use, into the limit of volumetric error.

3.2.5. If we turn to written standards that set out principles of specifications of laboratory volumetric glassware, e.g. the ISO standard [91], we will see that a run-to-run variation in use of volumetric apparatus is taken into account in the tolerance value. In particular, for glassware intended for delivery, where an error may be significant due to a variation in the draining technique, the limit of volumetric error is prescribed to be *not less than four times the experimental standard deviation obtained under repeatability conditions*.

Evidences available from experimental studies show that this holds true in laboratory practice. As an example, the data for one-mark pipettes, presented in the publication IV, demonstrate that the contribution of random error, expressed as 99.7% confidence interval, falls between 0.5 and 0.8 of the Class A tolerance value – in full agreement with the proportion ($\leq$ ¾) expected from the standard specification. Still smaller random variations are obtained when a higher level of accuracy is aimed at, specifically, in calibration services.

3.2.6. These findings testify that the capacity tolerance is the limit to allowable error in normal use of volumetric ware, not specifically in its calibration resulting in "calibration uncertainty" as the EURACHEM Guide states [5 (Appendix G)]. Of course, the procedures for proper use of volumetric glassware must be followed by qualified and motivated personnel. These procedures are laid down in the ISO standard [92] as well as applicable national standards, not to mention textbooks on quantitative chemical analysis which include chapters describing the subject in detail.

3.2.7. Compiling a cause and effect diagram is an effective means for analyzing uncertainty sources. Such a diagram constructed for a volumetric operation is presented in Fig. 4. Four main branches are depicted in the chart:

- *Procedure*: It is representative of the procedural contribution to the total uncertainty and incorporates the relevant factors.
- *Temperature effects*: Two different effects are taken into consideration – the variation of density of liquid with temperature and the change in the capacity of the vessel itself with the change of temperature. They act “in opposition”, with the former usually of much greater magnitude than the latter.
- *Calibration*: It includes its own subsidiary branches: *procedure* and *temperature*, the latter with two arrows as explained above, and some additional effects, specifically those associated with mass determination by weighing.
- *Physical properties of the liquid*: It is mainly concerned of the delivery processes and takes account of a difference in properties such as the viscosity and surface tension of the liquid being measured and water. For dilute aqueous solutions ordinarily employed these effects are small and they can be disregarded.

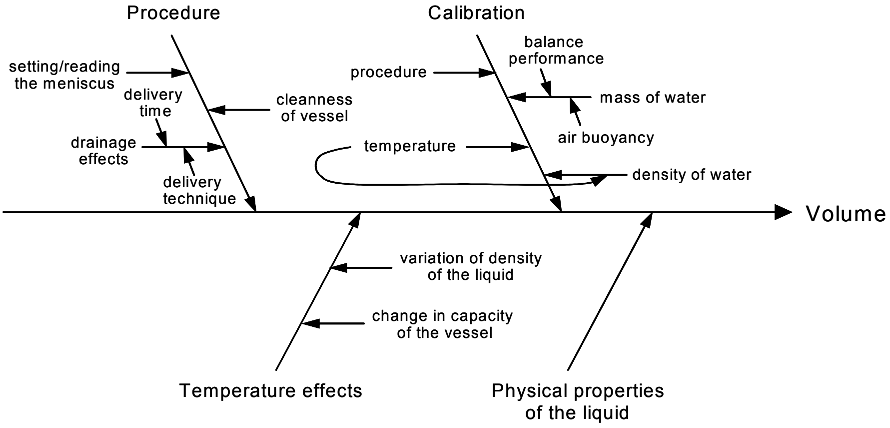


Figure 4. Cause and effect diagram for a volumetric operation

3.2.8. In a typical case where volumetric glassware is normally used in a laboratory, a practical way for the evaluation of measurement uncertainty is to use the tolerances from the specification. The influence factors relating to the upper branches, *Procedure* and *Calibration*, in the cause and effect diagram are covered in the stated tolerance. Only the bottom branches, namely, *Temperature effects*, should additionally be allowed for. With this supplementary contribution, the use of the capacity tolerance converted to the standard uncertainty is well justified.

3.2.9. In case where a high level of accuracy not attainable with the specified tolerances is required, or the manufacturer of the glassware cannot be confidently relied upon, or, finally, the volumetric glassware is misused or damaged, it is reasonable to assess the actual performance in the user's hand, carrying out a volumetric repeatability experiment. This is a calibration experiment in a way, based on the gravimetric determination of the actual volume contained or delivered. In this way, the volumetric apparatus factory calibrated becomes that in-house calibrated, and we will use the estimated true capacity instead of the nominal capacity and also the experimental standard deviation of the mean as a more specific estimate of calibration uncertainty than that derived from the tolerance.

3.2.10. An important point is that the uncertainties from both the measurement and calibration are estimated now in terms of the experimental standard deviation, so that there is no need for utilizing the manufacturer’s tolerances. Thus, we come to recognize that the specified tolerances and the estimated variability shall be used alternatively, not conjointly as the guide [5] recommends based on a misinterpretation of volumetric tolerance. Two different procedures can be referred to as the *tolerance-based* approach and the *actual performance-based* approach. In the first case, the nominal capacity and a tolerance-based uncertainty are taken as the measurement result while in the second, the estimated true capacity (of a specific volumetric apparatus) is taken, usually provided with much smaller uncertainty.

3.2.11. In Table 1 the full uncertainty budget for the two approaches is drawn up to comparing different contributions to the total uncertainty, which allows us to judge the efficiency and suitability one way or the other. The budget is exemplified by calculations and experimental data obtained for 100 ml class B volumetric flasks.

As is clear from the example, the resulting uncertainty is reduced substantially if it is based on the actual performance rather than on the specification. This is due to the small uncertainty contribution that the random variation actually introduces into the budget, and still smaller remaining contributions of calibration uncertainty. Also, a significant calibration bias (0.15 ml) is revealed. It does not follow herefrom yet that the analyst must necessarily be engaged in a performance exercise with his volumetric glassware unless it is justified by a sufficient reason as mentioned in 3.2.9.

Among other things it can be seen from the table that the uncertainty caused by the temperature variation (calculated with the normal assumption of fluctuations within ± 4°C about the mean room temperature) becomes dominating contribution in the actual performance approach. The inference to be made is that in-house calibration is unjustified when the temperature control in use of volumetric apparatus is lacking. It is unreasonable to make calibration with a high level of accuracy that is not required and not ensured in the measurement.

Table 1. Uncertainty budget for a volumetric operation – measurement of liquid volume with a volumetric flask

Uncertainty source	Standard uncertainty	
	Formula [a]	Example [b], ml
(1) The tolerance-based approach		
Procedure Calibration	$\left.\begin{matrix} \\ \\ \end{matrix}\right\} \quad \frac{\Delta_\text{v}}{\sqrt{6}}$	0.082
Temperature	$\frac{V \alpha \Delta t}{\sqrt{6}}$	0.034
Combined standard uncertainty		0.089
Expanded uncertainty ($k = 2$)		0.18
Result of a measurement		100.00 ± 0.18
(2) The actual performance-based approach		
Procedure	s	0.014
Calibration:		
-procedure	$s/\sqrt{n}$	0.0044
-balance performance	$\frac{1}{\rho_\text{w}}\sqrt{s_\text{b}^2 + 2\frac{\Delta_\text{nl}^2}{3}}$	0.00013
-density of water /temperature	$\frac{V}{\rho_\text{w}}\left(\frac{\text{d}\rho_\text{w}}{\text{d}t}\right)_\text{t}\frac{\Delta_t}{\sqrt{6}}$	0.0017
Temperature	$\frac{V \alpha \Delta t}{\sqrt{6}}$	0.034
Combined standard uncertainty		0.037
Expanded uncertainty ($k = 2$)		0.074
Result of a measurement		100.15 ± 0.07

a) In these formulas: Δ_v is the volumetric tolerance specified, V is the nominal capacity for a volumetric apparatus factory calibrated or the estimated true capacity for that in-house calibrated, α is the coefficient of cubical thermal expansion of the liquid measured, Δt is the limit of possible temperature variation about the mean working temperature, s is the standard deviation from the repeatability (calibration) experiment with the number of replicates equal to n, ρ_w is the density of water as calibrating liquid, s_b is the repeatability of the balance specified as the standard deviation, Δ_nl is the non-linearity of the balance specified as the maximum allowable deviation from the linear characteristic function, and Δ_t is the limit of possible temperature variation in calibration process.

b) As an example, the uncertainties for a 100 ml Class B volumetric flask were calculated based on the specification (1) and the calibration experiment (2). For details see the original publication IV

3.2.12. It is concluded that the capacity tolerance specified for volumetric apparatus can suitably be used in evaluating volumetric uncertainty, with no additional allowance needed except the temperature effects. Alternatively, the actual performance has to be estimated if necessary in a repeatability (calibration) experiment irrespective of the prescribed tolerance. This is two different procedures not to be combined in a single one. The original EURACHEM recommendation comprises an excess procedure, and there is little reason for replicating it in recent textbooks [93–95] designed for a large laboratory audience.

3.3. Treatment of measurement bias in relation to uncertainty estimation

The requirement on correction for recognized systematic effects is one of the central points of the measurement uncertainty methodology in the GUM [4 (3.2.4)]. The uncertainty components are combined as centered random variables, while strong systematic effects are to be corrected for. It is supposed that potential bias is investigated and, if statistically significant in comparison with the uncertainty on the observed difference, is corrected for in the measurement result. The uncertainty associated with the bias correction is then combined with other uncertainty components.

The problem of bias handling in analytical measurement has received much attention in recent literature [e.g. 24, 96–98], particularly, with regard to the necessity of treating uncorrected (even significant) bias where making correction is considered unpractical from technical or economical point of view.

As discussed in the recent paper [98], in those cases where (1) the bias is significant, (2) the cause of the bias is understood, (3) an accurate estimate of the effect is available, and (4) a useful reduction in uncertainty on applying the correction will occur, the correction for that bias is well justified for technical reason. And conversely, if none of the above criteria is hold, correction is unjustified and impossible.

There are, nevertheless, striking examples, in which the recommendations have been given quite opposite to those resulting from the analysis of the situations. One of the examples concerns the evaluation of secondary pH standards which are assigned a certain level of uncertainty in the pH traceability chain. The other one relates to common practice of chemical analysis in a laboratory. These examples that are both from authorities in their analytical fields are analyzed in subsequent sections 3.3.1 and 3.3.2, respectively.

3.3.1. Evaluation of secondary pH standards: accounting for liquid junction potential contribution

3.3.1.1. The requirement of metrological traceability of pH measurements implies that a hierarchical system is established linking working measurements to a primary standard and if needed to SI units, all links in the chain having stated uncertainties. At the highest level, the pH value of a buffer solution identified as a primary pH standard is determined with the lowest uncertainty (0.003 – 0.004 in pH units as expanded uncertainty[1]) from electrochemical cell without transference, the Harned cell, which constitutes the primary method of measurement. The pH values of other buffer solutions referred to as secondary pH standards are derived from the value of the primary standard by comparison in a cell with liquid junction, having greater uncertainty associated with the results. The secondary standards are used for calibration of cells involving glass electrodes and liquid junctions (routine pH instruments) at the working measurement level, providing finally the measurement results with even greater uncertainty (typically 0.02 – 0.03 for 2-point calibration).

3.3.1.2. The evaluation of pH of one buffer solution with respect to the other having a different composition is based on emf measurements of the differential cell containing the two solutions, S_1 and S_2, separated with a salt bridge:

$$\mathrm{Pt} \mid \mathrm{H_2} \mid \mathrm{S_2} \,¦\, \mathrm{KCl}\ (\geq 3.5\ \mathrm{mol\ dm^{-3}}) \,¦\, \mathrm{S_1} \mid \mathrm{H_2} \mid \mathrm{Pt} \qquad (3)$$

In this way, the pH of one of the solutions is expressed in terms of the other:

$$\mathrm{pH(S_2)} = \mathrm{pH(S_1)} - E_{\Delta\mathrm{pH}}/k + (E_{\mathrm{j2}} - E_{\mathrm{j1}})/k \qquad (4)$$

where the difference ($E_{\mathrm{j2}} - E_{\mathrm{j1}}$) is the residual liquid junction potential contribution to the cell emf and $k = (RT/F)\ln 10$ is the Nernst slope. A free-diffusion liquid junction implemented in a vertical capillary tube, which is known to have the best performance, is used in such measurements.

3.3.1.3. In the recent IUPAC Recommendations *Measurement of pH. Definition, standards, and procedures* [99], typical uncertainty budgets are presented for pH measurements at primary, secondary and working levels. Specifically, in the budget for secondary pH buffer using cell (3), the standard uncertainty due to RLJP is estimated as 0.006; this is the principal contribution in the budget. As a result, an unjustifiably high uncertainty level (0.015 as expanded uncertainty)

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[1] This is the intrinsic uncertainty in the measured pH value of a buffer solution without regard for additional uncertainties due to instability, possible contamination, and so on, which should be accounted for in the sample on the desk of the user.

has been established for secondary pH standards, approximately four times larger than for primary ones.

3.3.1.4. The standard uncertainty of 0.006 in [99] is derived from the limits ± 0.01 to possible values of the RLJP error, assuming that the distribution is rectangular. The limits have been estimated from the comparison of the values pH(SS) of several secondary buffer solutions with their values pH(PS) assigned by primary method. Such measurements, with the (1:1) phosphate buffer as a reference, demonstrate that the RLJP errors in eqn (4) are within ± 0.01 in intermediate pH range ($3 \leq \text{pH} \leq 11$) at 25 °C, as indicated in Fig. 2 in the IUPAC document [99].

This "statistical" approach to the assessment of the RLJP uncertainty is, however, in conflict with simple logic: it would be true only if it were unknown which pair of buffer solutions is compared in a particular measurement. But actually, we always know the nature of the buffer solutions we are measuring. For this reason such a global estimate of uncertainty caused by the liquid junction potential is unjustified, particularly, if reproducible junctions of the free-diffusion type are employed.

3.3.1.5. The diffusion potential that generally arises in a heteroionic junction between two electrolyte solutions displaces the value of secondary buffer solution from its "true" value, i.e. the value that would be obtained if measured by the primary method, in a certain direction. The magnitude of the displacement is determined by the composition of the solutions and the geometry of the junction. For this reason the RLJP error is principally systematic for a given pair of solutions, not random; this error should certainly be treated as bias in uncertainty estimation.

3.3.1.6. A direct evidence for the systematic character of this RLJP error can be seen from data on the differences $\delta\text{pH} = \text{pH(SS)} - \text{pH(PS)}$ for a specific system: phthalate / (1:1) phosphate buffer solutions. The data are presented in the original publication III, calculated from the results of several researchers over the period of nearly 30 years. The δpH values agree satisfactory: the mean value from the five different figures is estimated as 0.008 with the standard deviation of the mean as low as 0.001.

3.3.1.7. It is appropriate to use the δpH value so obtained to make a correction in the evaluation of a secondary standard buffer. All the criteria (i)–(iv) for bias correction [98] are met in this case. Then, the RLJP uncertainty contribution in the original budget is replaced by the uncertainty of the correction, equal to the estimated standard deviation of the mean δpH. As a result, we get the combined standard uncertainty of the secondary pH standard practically at the same level (0.003 in this case) as for the primary standard from which the former is derived. The significantly reduced intrinsic uncertainty in the pH value of the standard buffer solution (0.006 as expanded uncertainty) is eventually obtained.

The findings above may not be necessarily so good for other buffer systems because of the lack of sufficient data. Nevertheless, if a correction is not completely reliable at present, it might be refined as new data become available.

3.3.1.8. The level of uncertainty assigned to secondary buffer solutions is critical in the context of the problem of two pH scales, the multiple-standard and the single-standard scale, which was a source of a long-standing controversy among pH experts [100, 101]. With a high target uncertainty assigned to secondary standards, such as 0.015, the difference between pH values on the two scales becomes insignificant, and the problem of two scales seems to have been solved.

In actual fact, both of the “scales” are involved in pH, operating at different levels in the hierarchy. This is a characteristic feature of the pH traceability chain that bias emerges on going from the primary level to the secondary level as a result of a liquid-junction potential contribution in the differential cell. Although not frequent, there are similar examples of change in size of a unit in a traceability chain in metrology. For instance, this is the case in mass metrology where two concepts are in use, “true mass” and “apparent mass”, related to each other by a buoyancy correction.

3.3.1.9. The measurement and uncertainty relationships along the traceability chain in pH can be displayed schematically as shown in Fig. 4. In the diagram, the route of traceability is laid down with the transition from the value pH(PS) of a primary standard to the value pH(SS) of a secondary standard, followed by a correction equal in magnitude to the junction bias Δ_{j}, depicted by a bent arrow. The transition from the value $\mathrm{pH(SS)_{corr}}$ to the value pH(X) of an unknown sample is marked by $\Delta_{\mathrm{pH(X)}}$. At the working measurement level, the RLJP effects may be different in magnitude and sign, resulting in an additional uncertainty contribution shown as U_{j} in the diagram.

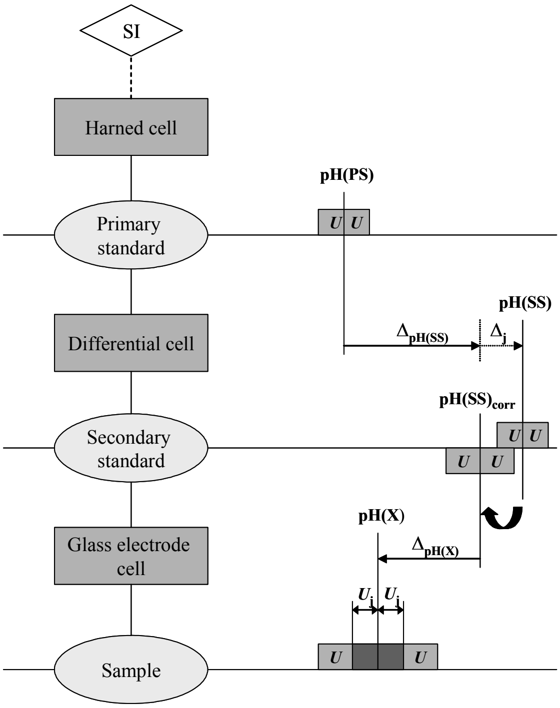

Figure 4. Traceability route with measurement and uncertainty relationships in pH

3.3.2. Run bias: do we really need to correct for it in chemical analysis?

3.3.2.1. A direct way for the bias / uncertainty estimation is applying the analytical procedure to an appropriate reference material. (Alternatively, the procedure under investigation and the reference procedure are applied in parallel to appropriate samples.) By comparison the mean of repeated measurements with the reference value, analytical bias can be detected and its significance assessed. It is generally accepted that intermediate precision ("within-laboratory reproducibility") conditions are preferred in the bias study, since more effects may be covered in this case than when working under repeatability conditions, and a more representative estimate of the uncertainty may be obtained.

3.3.2.2. It has been recently proposed [102] with regard to uncertainty estimation that analytical bias be determined under repeatability conditions, by a single analysis of a CRM in each analytical run. The authors [102] come to the conclusion that the best strategy is to always correct the results for "run bias", i.e. the observed deviation from the reference value of the CRM. When no correction is applied, the resulting uncertainty that is also estimated under repeatability conditions should be enlarged by an appropriate method to take that uncorrected bias into account.

3.3.2.3. The starting point in the issue of bias is the question whether the observed difference is statistically significant or not. Depending on which standard deviation is used in testing the significance, whether repeatability, or intermediate precision, or reproducibility, different conclusions may be reached. If the repeatability standard deviation is taken (root-sum-squared with the standard uncertainty of the reference value), the "run bias" has indeed a good chance to be found significant. The authors [102] insist just on this kind of testing, acknowledging that "bias is often hidden by the significance test being based on reproducibility data".

This is a question, answering which still separates the attitudes of a research analytical chemist and that engaged in providing analytical services. The authors [102] proceed from the belief that "…most analysts would consider that they primary objective is to obtain a test result that is as close to the true value of the measurand as possible". However, the objective of the analyst in analytical service, or analytical laboratory on the whole, is quite different – to produce a result the quality of which is in agreement with its intended use, that is, "fit for purpose". If the quest for reducing the measurement error does not emerge from fitness for purpose requirements, any effort to find what is "hidden" is unnecessary. It leads to a false conclusion that a significant bias occurs while in fact, using the (proper) measurement uncertainty criterion, it may not occur and usually does not.

3.3.2.4. If we look closer at what can be expected from the correction for "run bias", we will find that the outcome is not so inviting as it might seem at first sight. The benefit from cancelling bias will be largely "compensated" by additional uncertainty contributions introduced with the correction. This is particularly the case when a single analysis of the reference material is carried out, resulting in doubling the random error contribution in the uncertainty.

According to the "ladder of errors" model [61], the single test result x is expressed in terms of the true value μ, the repeatability error ε, and the corresponding contributions from run bias δ_{run}, laboratory bias δ_{lab}, and method bias δ_{meth} as follows:

$$x = \mu + \varepsilon + \delta_{\text{run}} + \delta_{\text{lab}} + \delta_{\text{meth}} . \tag{5}$$

Then, the combined uncertainty u of the result is given by [85]:

$$u = \sqrt{\sigma_{\varepsilon}^2 + \sigma_{\text{run}}^2 + \sigma_{\text{lab}}^2 + u_{\text{b}}^2} \tag{6}$$

where σ_{ε}^{2} is the variance in repeatability conditions, σ_{run}^{2} and σ_{lab}^{2} are the variances of the populations of run and laboratory bias respectively, and u_{b} is the standard uncertainty for the method bias. In the case of correction for "run bias", the δ_{run}, δ_{lab}, and δ_{meth} contributions in (5) cancel out, but the uncertainty u_{corr} of the corrected result will amount to

$$u_{\text{corr}} = \sqrt{2\sigma_{\varepsilon}^{2} + u_{\text{CRM}}^{2} + u_{\text{matr}}^{2}}\ . \qquad (7)$$

Here u_{CRM} is the uncertainty of the reference value attributed to the CRM, and u_{matr} is the uncertainty caused by a matrix (and also, analyte level) mismatch between the test material and the reference material, with the repeatability standard deviation σ_{ε} taken equal for the test and reference samples. Although it is difficult to directly compare the magnitudes of uncertainty components in (6) and (7), one may expect that u_{corr} is unlikely to fall far short of u.

3.3.2.5. The uncertainty of the result as given by eqn (6) involves the run effect at the second level of organization in analytical system, in the wake of the repeatability error. It is evidently not reasonable to think of this effect as a systematic error if the next effects, the laboratory bias and even method bias, are included into the budget just as uncertainty components. It is important to realize that measurement uncertainty expresses the quality of analytical results in a global sense, providing the basis for their comparability "in space" of different times, different laboratories, and different methods. From this point of view, the run-to-run comparability is not a factor to be allowed for by a corrective action.

3.3.2.6. Considering the technical criteria (i)–(iv) for bias correction (see Section 3.3 above), we conclude that the change to repeatability conditions in determining bias brings to the fact that criterion (i) would be more easily met, as the standard deviation used in the significance test is (artificially) decreased. Meeting criterion (ii) becomes problematic, since a "causeless" run bias component appears in the observed difference. The estimate of bias, derived from a single CRM analysis, cannot be sufficiently accurate, therefore criterion (iii) and, as a consequence, criterion (iv) tend to be not met. Thus, correction for "run bias" can hardly be justified. And the uncertainties that would include that bias, if the correction is not applied, would be different from run to run, which is impractical.

Other aspects of the problem are discussed in the original publication V.

4. CONCLUSIONS

1. The measurement uncertainty methodology does not mean discarding the classical insight about analytical data quality expressed in terms of accuracy and error. It seems quite appropriate to use the philosophy of the classical approach combined with the tools and practical methodology provided by the uncertainty approach. Thus, the measurement uncertainty can be viewed as a single-number index of accuracy, being constructed following definite rules.
2. The concept of "systematic" error, a peculiar random variable that is defined on a set of possible implementations of the measurement procedure, is particularly appropriate in chemical analysis. This error occurs here not only on a set of procedure implementations, as is the case in physical measurement, but also on a set of samples differing in their matrix composition within the scope of the procedure. Both these components of "systematic" error contribute to the uncertainty of the result of analysis.
3. There is a good reason to explicitly include the quality matter in the concept "analytical procedure", thus giving a determining aspect of the notion. Defined in this way, analytical procedure is a "specified procedure to follow which ensures obtaining the results of analysis with a known uncertainty". The fitness-for-purpose criterion built in the definition offers an advanced view on the problem of "method validation". It is the measurement uncertainty that needs paramount consideration among other performance characteristics in validation studies.
4. The rational strategy of the uncertainty estimation involves identification of relevant sources of uncertainty and taking these into account using first of all the "whole method" performance data (precision and trueness). Uncertainty components that are not covered or are inadequately coved by the overall performance study are then accounted for separately. The use of any performance information, at least the repeatability standard deviation, is essential.
5. When considering uncertainty components on an analytical procedure, one has to identify the components as resulting from individual (or combined) sources of uncertainty rather than from separate parts of the analytical system, which are difficult to be evaluated in isolation. The example of a multistep analytical procedure where the effect on one step is evaluated from the final measurement result illustrates this feature.
6. In the evaluation of volumetric uncertainty, the specified tolerances and the estimated variability shall be used alternatively, not conjointly as is commonly recommended. These two different procedures can be referred to as the tolerance-based approach and the actual performance-based approach. In one case, the nominal capacity and the tolerance-based uncertainty are

taken as the measurement result while in the second, the estimated true capacity is taken, usually provided with much smaller uncertainty.

7. Treatment of measurement bias still remains an intricate point in analytical measurement. Two examples that have been fully considered demonstrate that the decisions taken are based on misinterpretation rather than a sound reasoning. Particularly, the bias in pH values of secondary standard buffer solutions, for which all the criteria for correction are met, has been statistically treated as a component of uncertainty. On the contrary, the random variable, "run bias" occurred in an analytical determination, was interpreted as a bias to be corrected for.

5. REFERENCES

1. ISO/IEC 17025:2005, *General requirements for the competence of testing and calibration laboratories*, International Organization for Standardization.
2. ISO 15189:2003, *Medical laboratories – Particular requirements for quality and competence*, International Organization for Standardization.
3. ILAC-G17:2002, *Introducing the concept of uncertainty of measurement in testing in association with the application of the standard ISO/IEC 17025*, International Laboratory Accreditation Cooperation.
4. ISO/IEC Guide 98:1995, *Guide to the expression of uncertainty in measurement*, International Organization for Standardization.
5. EURACHEM / CITAC Guide CG 4: *Quantifying uncertainty in analytical measurement*, 2nd ed. Eurachem, 2000.
6. V. J. Barwick and S. L. R. Ellison, *Protocol for uncertainty evaluation from validation data*, Report LGC/VAM/1998/088, LGC Ltd., January 2000.
7. EA-4/16, *EA guidelines on the expression of uncertainty in quantitative testing*, European Co-operation for Accreditation, December 2003.
8. ISO/TS 21748:2004, *Guidance for the use of repeatability, reproducibility and trueness estimates in measurement uncertainty estimation*, International Organization for Standardization.
9. B. Magnusson, T. Näykki, H. Hovind and M. Krysell, *Handbook for calculation of measurement uncertainty in environmental laboratories*, 2nd ed. NORDTEST Technical Report 537, February 2004.
10. *Guide to the evaluation of measurement uncertainty for quantitative test results*, EUROLAB Technical Report 1/2006, August 2006.
11. *Measurement uncertainty revisited: Alternative approaches to uncertainty evaluation*, EUROLAB Technical Report 1/2007, March 2007.
12. E. Desimoni and B. Brunetti, *Annali di Chim.*, 2005, **95**(5), 265.
13. I. Taverniers, E. Van Bockstaele and M. De Loose, *Trends Anal. Chem.*, 2004, **23**(7), 480.
14. Second EURACHEM Workshop: *Measurement uncertainty in chemical analysis: Current practice and future directions*. Berlin, 29–30 September 1997.
15. Third EURACHEM Workshop in co-operation with IAEA: *Efficient methodology for the evaluation of uncertainty in analytical chemistry; Implementing the ISO guide*. Helsinki, (13)14–15 June 1999.
16. *Measurement uncertainty in chemical analysis*, P. De Bièvre and H. Günzler (Eds.). Berlin, etc.: Springer, 2003.
17. S. K. Kimothi, *Uncertainty of measurement: Physical and chemical metrology: Impact and analysis*. Milwaukee, Wis.: ASQ Press, 2002.
18. R. Visser, *Accred. Qual. Assur.*, 2004, **9**(11/12), 717.
19. A. M. H. van der Veen, *Accred. Qual. Assur.*, 2003, **8**(6), 300.
20. W. Horwitz and R. Albert, *Analyst*, 1997, **122**(6), 615.
21. W. Horwitz, *J. AOAC Int.*, 1998, **81**(4), 785.
22. A. B. Blank, *J. Anal. Chem.*, 2005, **60**(12), 1173.
23. A. N. Smagunova, *Zavod. Lab. Diagn. Mater.*, 2007, **73**(1), 116 (in Russian).
24. W. Hässelbarth, *Accred. Qual. Assur.*, 2004, **9**(8), 509.
25. B. King, *Metrologia*, 1997, **34**(1), 41.

26. E. Hund, D. L. Massart and J. Smeyers-Verbeke, *Trends Anal. Chem.*, 2001, **20**(8), 394.
27. ISO/IEC Guide 99:2007, *International vocabulary of metrology – Basic and general concepts and associated terms* (VIM, 3rd edn), International Organization for Standardization.
28. *International vocabulary of basic and general terms in metrology* (VIM), 2st edn, International Organization for Standardization, 1993.
29. *International vocabulary of basic and general terms in metrology* (VIM), 1st edn, International Organization for Standardization, 1984.
30. P. J. Campion, J. E. Burns and A. Williams, *A code of practice for the detailed statement of accuracy*, National Physical Laboratory. London: Her Majesty's Stationery Office, 1973.
31. C. Eisenhart, *Science*, 1968, **160**, 1201.
32. A. R. Colclough, *J. Res. Nat. Bur. Stand.*, 1987, **92**(3), 167.
33. H.-J. Martens, *Feingerätetechnik*, 1981, **30**(10), 443; **30**(11), 514.
34. R. B. Abernethy and B. Ringhiser, *The history and statistical development of the new ASME-SAE-AIAA-ISO measurement uncertainty methodology*. AIAA/SAE/ASME/ASEE 21st Joint Propulsion Conference. Monterey California, July 8–10, 1985.
35. H. W. Coleman and W. G. Steel, Jr, *Experimentation and uncertainty analysis for engineers*. New York, etc.: Wiley, 1989.
36. R. H. Dieck, *Measurement uncertainty: Methods and applications: A guide to estimating and understanding the accuracy of test and experimental data*, 2nd edn, Research Triangle Park. NC: Instrument Society of America, 1997.
37. L. A. Currie, *Sources of error and the approach to accuracy in analytical chemistry*, Treatise on analytical chemistry, I. M. Kolthoff and P. J. Elving (Eds.), Part I, Theory and practice, V.1, 2nd edn. New York, etc.: Wiley, 1978. P. 95.
38. *A.S.T.M. Manual on presentation of data: Supplement A – Presenting ± limits of uncertainty of an observed average*. Philadelphia: American Society for Testing Materials, 1935.
39. R. F. Moran, *Ind. Eng. Chem. Anal. Ed.*, 1943, **15**(6), 361.
40. M. G. Mellon, *Quantitative analysis: Methods of separation and measurement*. New York: Thomas Y. Crowell Comp., 1955. P. 375.
41. J. K. Taylor, *Quality assurance of chemical measurements*. Chelsea, MI: Lewis Publishers, 1987.
42. E. F. McFarren, R. J. Lisha and J. H. Parker, *Anal. Chem.*, 1970, **42**(3), 358.
43. J. O. Westgard, R. N. Carey and S. Wold, *Clin. Chem.*, 1974, **20**(7), 825.
44. R. Wolters and G. Kateman, *Anal. Chim. Acta*, 1988, **207**, 111.
45. M. Valcárcel and A. Ríos, *Anal. Chem.*, 1993, **65**(18), 781A.
46. R. L. Kadis, *J. Anal. Chem.*, 2007, **62**(6), 506.
47. C. Eisenhart, *J. Res. Nat. Bur. Stand.*, 1963, **67C**(2), 161.
48. W. J. Youden, *Accuracy and precision: Evaluation and interpretation of analytical data*, Treatise on analytical chemistry, I. M. Kolthoff and P. J. Elving (Eds.), Part 1, Theory and practice, Vol. 1, 1st edn. New York, etc.: Wiley, 1959. P. 47.
49. J. Mandel, *Accuracy and precision: Evaluation and interpretation of analytical results*, Treatise on analytical chemistry, I. M. Kolthoff and P. J. Elving (Eds.), Part 1, Theory and practice, Vol. 1, 2nd edn. New York, etc.: Wiley, 1978. P. 243.

50. G. T. Wernimont, *Use of statistics to develop and evaluate analytical methods*. Gaithersburg MD: AOAC International, 1985.
51. IUPAC Protocol for the design, conduct and interpretation of method-performance studies, *Pure Appl. Chem.*, 1995, **67**(2), 331.
52. ISO 5725:1994, *Accuracy (trueness and precision) of measurement methods and results*, Parts 1–6 (Part 5 is 1998), International Organization for Standardization.
53. C. Ehrlich, R. Dybkaer and W. Wöger, *Accred. Qual. Assur.*, 2007, **12**(3/4), 201.
54. V. M. Sviridenko, *Measur. Techn.*, 1971, **14**(5), 667.
55. L. Mari, *Measurement*, 2005, **38**(4), 259.
56. J. Michell, *History and philosophy of measurement: A realist view*, Proc. 10th IMEKO TC7 International symposium on advances of measurement science. St. Petersburg, June 30–July 2, 2004.
57. P. De Bièvre, *Accred. Qual. Assur.*, 2000, **5**(7), 265.
58. P. De Bièvre, *Accred. Qual. Assur.*, 2006, **10**(12), 645.
59. *Terminology – the key to understanding analytical science, Part 1: Accuracy, precision und uncertainty*, AMC Technical Brief, 13, 2003.
60. V. V. Nalimov, *Application of mathematical statistics to chemical analysis*. Oxford: Pergamon, 1963. P. 6.
61. M. Thompson, *Analyst*, 2000, **125**(11), 2020.
62. T. V. Korneeva, *Explanatory dictionary on metrology, measurement techniques, and quality control*. Moscow: Russkii Yazyk, 1990 (in Russian).
63. R. Kaiser, *Z. Anal. Chem.*, 1971, **256**(1), 1.
64. V. I. Paneva, N. A. Makulov and O. B. Korotkina, *Development and attestation of procedures for the quantitative analysis of substances and materials*. Moscow: Mashinostroenie, 1987 (in Russian).
65. B. Ya. Kaplan, L. N. Filimonov and I. A. Maiorov, *Metrology of the analytical monitoring of production in nonferrous metallurgy*. Moscow: Metallurgiya, 1989 (in Russian).
66. S. A. Wagner, *PTB Mitteilungen*, 1979, **89**(2), 83.
67. W. Wöger, *Randomization of systematic errors and its consequences for the evaluation of measurements*, Quantum metrology and fundamental physical constants, P. H. Cutler and A. A. Lucas (Eds.). New York and London: Plenum, 1983.
68. M. A. Zemel'man, *Metrological basics for technical measurements*. Moscow: Izd. Standartov, 1991 (in Russian).
69. N. A. Makulov, *Zavod. Lab.*, 1976, **42**(12), 1457 (in Russian).
70. A. Parczewski and A. Rokosz, *Chem. Analit.*, 1978, **23**, 225.
71. T. B. Crumpler and J. H. Yoe, *Chemical computations and errors*. New York: Wiley; London: Chapman and Hall, 1940. Ch. VII.
72. K. Doerffel, *Fresenius. J. Anal. Chem.*, 1998, **361**(5), 393.
73. J. M. Cameron, *J. Qual. Technol.*, 1976, **8**(1), 53.
74. P. E. Pontius, *Notes on the fundamentals of measurement and measurement as a production process*, NBSIR 74-545. Washington, D. C.: National Bureau of Standards, 1974.
75. W. J. Youden, *Anal. Chem.*, 1960, **32**(13), 23A.
76. W. J. Youden, *Mat. Res. Stand.*, 1961, **1**(4), 268.
77. J. K. Taylor, *Anal. Chem.*, 1983, **55**(6), 600A, 608A.

78. M. Parkany, *J AOAC*, 1986, **69**(3), 396.
79. D. Holcombe, *Accred. Qual. Assur.*, 1999, **4**(12), 525.
80. *Validation of analytical procedures: Text and methodology*, Q2 (R1), ICH Harmonized Tripartite Guideline, International Conference on Harmonization of Technical Requirements for Registration of Pharmaceuticals for Human Use, 1994. (http://www.ich.org/LOB/media/MEDIA417.pdf)
81. EURACHEM Guide: *The fitness for purpose of analytical methods: A laboratory guide to method validation and related topics*. LGC (Teddington) Ltd, 1998.
82. IUPAC Harmonized guidelines for single-laboratory validation of methods of analysis, *Pure Appl. Chem.*, 2002, **74**(5), 835.
83. M. Thompson and R. Wood, *Accred. Qual. Assur.*, 2006, **10**(9), 471.
84. D. L. Massart, A. Dijkstra and L. Kaufman, *Evaluation and optimization of laboratory methods and analytical procedures*. Amsterdam, etc.: Elsevier, 1978. Ch. 28 and 29.
85. Analytical Methods Committee, *Analyst*, 1995, **120**(9), 2303.
86. S. L. R. Ellison, *Accred. Qual. Assur.*, 1998, **3**(3), 95.
87. S. L. R. Ellison and V. J. Barwick, *Accred. Qual. Assur.*, 1998, **3**(3), 101.
88. S. L. R. Ellison and V. J. Barwick, *Analyst*, 1998, **123**(6), 1387.
89. *Quantifying uncertainty in analytical measurement*, Version 6. Eurachem, 1995.
90. K. Doerffel, *Statistik in der analytischen Chemie,* 5. Aufl. Leipzig: Deutscher Verlag für Grundstoffindustrie, 1990.
91. ISO 384:1978, *Laboratory glassware − Principles of design and construction of volumetric glassware*, International Organization for Standardization.
92. ISO 4787:1984, *Laboratory glassware − Volumetric glassware − Methods for use and testing of capacity*, International Organization for Standardization.
93. I. Mueller-Harvey and R. M. Baker, *Chemical analysis in the laboratory: A basic guide*. Cambridge: RSC, 2002. Ch. 4.
94. E. Mullins, *Statistics for the quality control chemistry laboratory*. Cambridge: RSC, 2003. Ch. 8.
95. D. B. Hibbert, *Quality assurance for the analytical chemistry laboratory*. Oxford, etc.: Oxford University Press, 2007. Ch. 6.
96. A. Maroto, R. Boqué, J. Riu and F. X. Rius, *Accred. Qual. Assur.*, 2002, **7**(3), 90.
97. V. Synek, *Talanta*, 2005, **65**(4), 829.
98. B. Magnusson and S. L. R. Ellison, *Anal. Bioanal. Chem.*, 2008, **390**(1), 201.
99. R. P. Buck, S. Rondinini, A. K. Covington, F. G. K. Baucke, M. F. Camoes, M. J. T. Milton, T. Mussini, R. Naumann, K. W. Pratt, P. Spitzer and G. S. Wilson, *Pure Appl. Chem.*, 2002, **74**(11), 2169.
100. A. K. Covington, *Anal. Chim. Acta*, 1981, **127**, 1.
101. W. F. Koch, *Chem. Eng. News*, 1997, **75**(42), 6.
102. G. E. O'Donnell and D. B. Hibbert, *Analyst*, 2005, **130**(5), 721.

6. SUMMARY

The thesis is concerned with theoretical and practical problems of measurement uncertainty evaluation in analytical chemistry.

The first part focuses on key aspects of measurement uncertainty in its relation to other metrology and quality concepts, first of all, accuracy and error which are central to classical approach to measurement. The subdivision of measurement errors into random and systematic is discussed in detail, and the concept of "systematic" error is highlighted as being of particular importance in analytical measurements. The integration of quality aspects into common analytical concepts is demonstrated with a definition of "analytical procedure" proposed. The recommended approaches to uncertainty evaluation in analytical chemistry are compared and a rational strategy is formulated.

The second part of the work analyses some points of misinterpretation in the evaluation of analytical uncertainties and provides correct solutions to the problems that have not been treated properly. The following issues are covered: quantification of uncertainty and correction factor in a multistep analytical procedure, evaluation of uncertainty in volumetric operations, accounting for liquid junction potential contribution in the evaluation of pH measurement standards, and treatment of run bias in routine analytical determinations. The latter two issues address the problem of bias handling which still remains an intricate point in analytical measurement.

The thesis includes the conclusions reached, the references, and 6 original publications.

7. SUMMARY IN ESTONIAN

Mõõtemääramatuse hindamine analüütilises keemias: seotud mõisted ja väärtõlgendused

Käesolev dissertatsioon keskendub mõõtemääramatuse hindamise teoreetilistele ja praktilistele aspektidele analüütilises keemias.

Töö esimene osa on pühendatud mõõtemääramatuse kontseptuaalsetele aspektidele ja seostele teiste metroloogia ja kvaliteedi hindamise lähenemisviisidega, eeskätt mõistetega täpsus ja viga. Detailselt käsitletakse mõõtevigade klassifikatsiooni süstemaatilisteks ja juhuslikeks ja rõhutatakse "süstemaatilise" vea iseäranis kõrget tähtsust keemilises analüüsis. Pakutakse mõiste "analüüsimetoodika" täpne definitsioon ja demonstreeritakse selle olulisust analüüsitulemuste kvaliteeditagamises. Võrreldakse analüütilises keemias mõõtemääramatuse hindamiseks välja pakutud lähenemisviise ja formuleeritakse ratsionaalne strateegia mõõtemääramatuse hindamiseks.

Töö teises osas käsitletakse mitmeid kirjanduses ilmnevaid väärtõlgendusi mõõtemääramatuse hindamisel analüütilises keemias ja pakutakse neile korrektsed lahendused. Vaatluse all on järgmised probleemid: mõõtemääramatuse hindamine ja korrektsioonifaktorid paljuetapilistes analüüsimetoodikates, mõõtemääramatuse hindamine vedeliku mahu mõõtmise juures, difusioonipotentsiaalist tuleneva määramatuse hindamine pH standardlahuste väärtuste määramatuse juures ja päevasisese süstemaatilise vea arvestamine rutiintaseme analüütilises keemias. Kaks viimast probleemi on olulise ja komplitseeritud probleemide ringi – süstemaatiliste efektide arvestamine analüütilises keemias – erijuhud.

Dissertatsiooni lõpetavad järeldused, viited kirjandusele ja 6 algupärast teadusartiklit.

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PUBLICATIONS

I

Reproduced with kind permission from Springer Science + Business Media:
Accreditation and Quality Assurance, vol. 3, 1998, pages 237–241.
Evaluating uncertainty
in analytical measurements: the pursuit of correctness
Rouvim Kadis

II

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Accreditation and Quality Assurance, vol. 7, 2002, pages 294–298.
Analytical procedure in terms
of measurement (quality) assurance
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III

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Analytical and Bioanalytical Chemistry. 2002, vol. 374, pages 817–823.
Secondary pH standards and their uncertainty
in the context of the problem of two pH scales
Rouvim Kadis

IV

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Evaluation of measurement uncertainty in volumetric operations: the tolerance-based approach and the actual performance-based approach, pages 167–173.

V

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The Analyst, vol. 132, 2007, pages 1272–1274.

Do we really need to account for run bias when producing analytical results with stated uncertainty? Comment on ‘Treatment of bias in estimating measurement uncertainty’ by G. E. O’Donnell and D. B. Hibbert
Rouvim Kadis

VI

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Journal of Analytical Chemistry, 2008, vol. 63, pages 95–100.
Measurement uncertainty and chemical analysis
R. L. Kadis

CURRICULUM VITAE

Rouvim Kadis

Born:	January 3, 1948. Leningrad (now St. Petersburg), Russia
Citizenship:	Russian
Marital status:	married, son
Address:	D. I. Mendeleyev Institute for Metrology (VNIIM) 19 Moskovsky pr, 190005 St. Petersburg, Russia Phone: (812)379-7988 E-mail: rkadis@b10.vniim.ru, rkadis@mail.rcom.ru

Education

1965–1971	Lensoviet Technological Institute, Leningrad, Physico-chemical faculty, Chemical technology engineer 1971 (honours degree)
2007–present	University of Tartu, Faculty of science and technology, PhD student

Professional employment and retraining

1971–1975	Analytical chemical laboratory, All-Union Research Institute for Mechanical Treatment of Minerals, Leningrad, research engineer
1975–1992	Laboratory for physico-chemical methods, All-Union Research Technological Institute for Antibiotics and Medical Enzymes, Leningrad, researcher
1992–present	Laboratory of state measurement standards in analytical measurements, D. I. Mendeleyev Institute for Metrology, St. Petersburg, researcher, principal expert
2001	Department 3.1 Metrology in chemistry, Physikalisch-Technische Bundesanstalt, Braunschweig, Germany, visiting scientist

Major publications

№	Title	Author(s)	Publication details
1	Measurement uncertainty and chemical analysis	Kadis R.L.	*Journal of Analytical Chemistry*, 2008, 63(1), 95–100.
2	Do we really need to account for run bias when producing analytical results with stated uncertainty?	Kadis R.	*The Analyst*, 2007, 132(12), 1272–1274
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4	Metrological and statistical meaning of the concept "accuracy" in chemical analysis. ISO 5725, indexes of accuracy, and measurement uncertainty	Kadis R.L.	*Zavodskaya Laboratoriya. Diagnostika Materialov,* 2006, **72**(2), 53–60 (in Russian)
5	Metrological and statistical meaning of the concept "accuracy" in chemical analysis. Accuracy, true value, and accepted reference value	Kadis R.L.	*Zavodskaya Laboratoriya. Diagnostika Materialov,* 2005, 71(12), 53–59 (in Russian)
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8	Analytical procedure in terms of measurement (quality) assurance	Kadis R.	*Accreditation and Quality Assurance,* 2002, **7**(7), 294–298
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10	Metrological language in chemistry: New proposals versus "good old" terms	Kadis R.	*Accreditation and Quality Assurance,* 1999, **4**(4), 153–154
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CURRICULUM VITAE

Rouvim Kadis

Sünniaeg ja -koht:	3. jaanuar, 1948. Leningrad, Venemaa
Kodakondsus:	Venemaa
Perekonnaseis:	Abielus, üks poeg
Aadress:	Ülevenemaaline D. I. Mendelejevi nimeline Metroloogia instituut (VNIIM), 19 Moskovski pr, 190005 St. Petersburg, Venemaa Tel: (812) 323-9640, 379-7988 E-mail: rkadis@b10.vniim.ru, rkadis@mail.rcom.ru

Haridus

1965–1971	Lensovet'i nimeline Tehnoloogiainstituut, Leningrad, Füüsika-keemiateaduskond, Insener-keemiatehnoloog 1971 (kiitusega)
2007–	Tartu Ülikool, Loodus- ja tehnoloogiateaduskond, doktorant

Teenistuskäik

1971–1975	Maavarade mehaanilise töötlemise üleliiduline teadus-uuringute instituut, Leningrad, Keemilise analüüsi labor, insener-teadur
1975–1992	Üleliiduline meditsiiniliste antibiootikumide ja fermentide teadusuuringute ja tehnoloogia instituut, Leningrad, nooremteadur
1992–	Ülevenemaaline D. I. Mendelejevi nimeline Metroloogia Instituut, St. Petersburg, Füsiko-keemiliste mõõtmiste osakond, teadur, peaspetsialist
2001	Physikalisch-Technische Bundesanstalt, Braunschweig, Saksamaa, Osakond 3.1 Keemiametroloogia, külalisteadur

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