codex alimentarius commission

FOOD AND AGRICULTURE ORGANIZATION OF THE UNITED NATIONS

WORLD HEALTH ORGANIZATION

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Agenda Item 2 **CX/MAS 07/28/2-Add.2**

JOINT FAO/WHO FOOD STANDARDS PROGRAMME

CODEX COMMITTEE ON METHODS OF ANALYSIS AND SAMPLING
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MATTERS REFERRED TO THE COMMITTEE BY THE CODEX ALIMENTARIUS COMMISSION AND OTHER CODEX COMMITTEES

GUIDANCE ON MEASUREMENT UNCERTAINTY

(prepared by The United Kingdom)

BACKGROUND

The provisions on "The Use of Analytical Results: Sampling Plans, Relationship between the Analytical Results, the Measurement Uncertainty, Recovery Factors and Provisions in Codex Standards" were adopted by the Commission at its 29th Session (July 2006) (ALINORM 06/29/41, paras 33-34 and Appendix III). The actual text to be included in the Procedural Manual is given as part of the attached guidance paper.

The Delegation of Thailand, supported by other delegations, expressed its concerns with the provisions for measurement uncertainty and pointed out and that if each Commodity Committee had the possibility to decide how to address measurement uncertainty, this would lead to inconsistency throughout Codex and therefore clear guidance should be provided on the allowance for measurement uncertainty.

The Delegation therefore proposed to defer the adoption of the provisions on the Use of Analytical Results until such guidance had been developed.

After some discussion, the Commission agreed to adopt the recommendations for inclusion in the Procedural Manual as proposed and to refer to the Committee on Methods of Analysis and Sampling the request made by some delegations for further guidance in order to address measurement uncertainty

It in order to aid the development of such guidance that this paper has been prepared.

DISCUSSION

Measurement uncertainty has been a contentious issue for analytical chemists for many years. It has been addressed in a number of papers. In the majority of cases these papers have concentrated on how measurement uncertainty is to be estimated. It is only recently that there have been discussions on how measurement uncertainty is to be used.

This draft Guidance explain the situation and are intended to draw together the various developments in the area which have directly affected the Codex Alimentarius Commission. It is written in the form of "questions and answers".

Developments which are or which will have to be drawn together include the CAC Guidelines on Measurement Uncertainty, the section in the CAC Procedural Manual on "The use of analytical results: sampling plans, relationship between the analytical results, the measurement uncertainty, recovery factors and provisions in codex standards", Codex terminology definitions and those for the dispute situation.

RECOMMENDATIONS

It is recommended that:

- the Committee discusses whether guidance should be developed as requested by the delegation of Thailand at the 2006 Session of the Codex Alimentarius Commission
- whether, if it is agreed that such guidance should be developed, the attached draft could aid its development
- whether there are additional questions that should be addressed.

GUIDANCE ON MEASUREMENT UNCERTAINTY

INTRODUCTION

It has been agreed by the Codex Alimentarius Commission that the text given in Part A of this Guidance is to be included in the Procedural Manual. This makes extensive use of the concept of "Measurement Uncertainty". In particular it stresses that measurement uncertainty is to be taken into account when discussing limits and their compliance.

Previously Codex has adopted Guidelines On Measurement Uncertainty (CAC/GL 54-2004). These are given at Part B.

Nevertheless, the estimation of measurement uncertainty and its subsequent use is subject to misunderstanding amongst some delegates to the Codex Committee on Methods of Analysis and Sampling and to the Codex Alimentarius Commission. This guidance has been developed to help its appreciation.

It has been given in the form of questions and answers.

1. What is Measurement Uncertainty?

Analytical results are variable. Most quantitative analytical results take the form of "a ± 2u" or "a ± U" where "a" is the best estimate of the true value of the concentration of the measurand (the analytical result) and "u" is the standard uncertainty and "U" (equal to 2u) is the expanded uncertainty. The range "a ± 2u" represents a 95% level of confidence in which the true value would be found. The value of "U" or "2u" is the value which is normally used and reported by analysts, normally referred to as "measurement uncertainty" and may be estimated in a number of different ways.

In food analysis it is the 95% probability (i.e. 2u) which is used to calculate the expanded uncertainty. In other sectors may specify a different probability.

Thus measurement uncertainty may be regarded as the variability around the reported results which is quantified as the value "U" when considering the expanded uncertainty and within which the "true" result should lie.

2. Does Measurement Uncertainty Apply to both Sampling and Analysis?

Measurement uncertainty applies to the whole measurement process. For analysts only "analytical" measurement uncertainty has been considered but it is now increasingly being recognised that the whole system must be considered, and so "sampling" measurement uncertainty is gaining an increasing importance (see CX/MAS 07/28/11, February 2007). However, this guidance only considers "analysis".

3. What is the Relationship between Measurement Uncertainty, the Analytical Result and the Method Used to Obtain the Result?

It is the estimation of the measurement uncertainty associated with an analytical result that is important. Measurement uncertainty is not associated with a method, but the values that are obtained in the validation of a method may be used to estimate the uncertainty of a result in some situations. This differentiation between "result" and "validated method" is frequently not appreciated and so causes some confusion. It does mean that different laboratories, even if using the same (validated) method on the "same" sample may report different measurement uncertainties. This is to be expected. As a consequence precision values for a validated method (the repeatability and reproducibility values) cannot to be taken to be the measurement uncertainty without qualification. In particular additional factors such as bias, matrix effect, competence of laboratory must be considered. These are commented on in further detail in Parts F and H.

4. Measurement Uncertainty and ISO/IEC 17025:2005

The estimation of measurement uncertainty is an integral part of the accreditation process. The 17025 Standard states that measurement uncertainty must be estimated and then made available to the customer if requested by the customer. The Codex Alimentarius Commission has developed Guidelines which require laboratories involved in the import/export of foods to be accredited.

5. Use of Measurement Uncertainty and Definition of a Dispute Situation

A dispute will arise when considering a Codex specification, which is a maximum value, if:

- the export certificate states that the analytical result to which its associated measurement uncertainty is then added is less than the Codex specification (i.e. "x + U" < L, where x is the reported analytical result, U is the expanded uncertainty and L is the Codex specification, which is a maximum limit) and so the sample meets the Codex specification, and

- the import certificate states that the analytical results to which its associated measurement uncertainty is then deducted is still greater than the Codex specification (i.e. "x – U" > L, where x is the reported analytical result, U is the expanded uncertainty and L is the Codex specification, which is a maximum limit) and so the sample does not meet the Codex specification[1].

This assumes that the laboratory at importation will deduct the measurement uncertainty, as implied in Part A of this guidance. If the value after deduction is still greater than the specification, then it may be stated, ***beyond reasonable doubt***, that the sample is not compliant with the specification.

6. Values of Measurement Uncertainty Estimations

There is concern that some laboratories underestimate the size of their uncertainties and report unrealistically small uncertainties to their customers.

For chemical analyses, using the results from collaborative trials, it would not be unreasonable to anticipate that the (expanded) uncertainties reported by laboratories would be of the following orders:

[1] Suggested UK definition in comment on the dispute situation paper.

Concentration	Expanded Uncertainty	Range of Acceptable Concentrations*
100g/100g	4%	96 to 104g/100g
10g/100g	5%	9.5 to 10.5g/100g
1g/100g	8%	0.92 to 1.08g/100g
1g/kg	11%	0.89 to 1.11g/kg
100mg/kg	16%	84 to 116mg/kg
10mg/kg	22%	7.8 to 12.2mg/kg
1mg/kg	32%	0.68 to 1.32mg/kg
< 100µg/kg	44%	56 to 144µg/kg

* this effectively means that values falling within these ranges may be regarded as being of the same analytical population.

For microbiological analyses, where it is frequently stated that results within the range of +/- 0.5 log units are acceptable, then the range of actual counts that this equates to is frequently much larger than customers of analytical data appreciate (or require).

It would be expected that the reported measurement uncertainties by all laboratories would not significantly exceed the value of the estimated from the reproducibility standard deviation (S_R) at the concentration of interest if the laboratory is in "analytical control".

7. Procedures for Estimating Measurement Uncertainty

There are many procedures available for estimating the measurement uncertainty of a result. Some of the more common of these are outlined in Parts C to J.

However, within Codex there are now a number of formal quality assurance measures which have to be implemented by control laboratories. In particular, such laboratories have to be:

- accredited to an Internationally recognised Standard (now with ISO/IEC 17025 Standard); such accreditation is aided by the use of internal quality control procedures,
- participate in proficiency schemes, and
- use validated methods.

It is essential that the information provided as a result of these requirements being implemented is used by laboratories when estimating their measurement uncertainties in order to avoid unnecessary work being carried out by laboratories. In Codex, where there is a high emphasis being placed on the use of "fully validated" methods of analysis, i.e. methods which have been validated through collaborative trials, information obtained from such trials can be used in many situations.

In addition information derived from internal quality control procedures may also be used to estimate uncertainties in some situations.

For the analyst it is important that no unnecessary duplication of existing work is undertaken.

Laboratories may use information derived from the following procedures to aid their estimation of the uncertainty of measurement of their results. These are outlined in Parts C to G of this guidance:

Part C ISO Guide to the expression of uncertainty in measurement

Part D EURACHEM Guide to quantifying uncertainty in analytical measurement – introduction

Part E	EURACHEM Guide to quantifying uncertainty in analytical measurement – component-by-component approach
Part F	EURACHEM Guide to quantifying uncertainty in analytical measurement – use of collaborative trial data
Part G	Use of collaborative trial: data – ISO 5725 critical differences
Part H	ISO TS 21748 – Guide To The Use Of Repeatability, Reproducibility And Trueness Estimates In Measurement Uncertainty Estimation
Part I	Concept Set By Commission Decision 2002/657/EC Implementing Council Directive 96/23/EC Concerning The Performance Of Analytical Methods And The Interpretation Of Results
Part J	An AOAC International Commentary
Part K	Internal quality control approach
Part L	NMKL (Nordic Committee on Food Analysis) approach

This information is outlined in the following sections of the Annex.

There is no "hierarchy" of procedures given to the sections. They are considered to be equally valid. However, the procedure that an individual laboratory uses will be considered appropriate by its Accreditation Agency as part of its 17025 accreditation.

It is recognised that further procedures for the estimation of measurement uncertainty are being developed, and that, in this evolving situation, further recommendations will be made as to acceptable procedures. It is anticipated that procedures based on results obtained from participation in proficiency testing schemes, as an example, will be developed.

8. Useful References

A number of references are given in Part M of this document. These are general references and do need up-dating.

PART A: TEXT (TO BE INCLUDED IN THE CODEX PROCEDURAL MANUAL AT THE END OF THE SECTIONS ON METHODS OF ANALYSIS AND SAMPLING IN THE *GUIDELINES FOR THE INCLUSION OF SPECIFIC PROVISIONS IN CODEX STANDARDS AND RELATED TEXTS)*

THE USE OF ANALYTICAL RESULTS: SAMPLING PLANS, RELATIONSHIP BETWEEN THE ANALYTICAL RESULTS, THE MEASUREMENT UNCERTAINTY, RECOVERY FACTORS AND PROVISIONS IN CODEX STANDARDS

ISSUES INVOLVED

There are a number of analytical and sampling considerations which prevent the uniform implementation of legislative standards. In particular, different approaches may be taken regarding sampling procedures, the use of measurement uncertainty and recovery corrections.

At present there is no official guidance on how to interpret analytical results in the framework of Codex. Significantly different decisions may be taken after analysis of the "same sample". For example some countries use an "every-item-must-comply" sampling regime, others use an "average of a lot" regime, some deduct the measurement uncertainty associated with the result, others do not, some countries correct analytical results for recovery, others do not. This interpretation may also be affected by the number of significant figures included in any commodity specification.

It is essential that analytical results be are interpreted in the same way if there is to be harmonisation in the framework of Codex.

It is stressed that this is not an analysis or sampling problem as such but an administrative problem which has been highlighted as the result of recent activities in the analytical sector, most notably the development of International Guidelines on the Use of Recovery Factors when Reporting Analytical Results and various Guides prepared dealing with Measurement Uncertainty.

RECOMMENDATIONS

It is recommended that when a Codex Commodity Committee discusses and agrees on a commodity specification and the analytical methods concerned, it states the following information in the Codex Standard:

1. Sampling Plans

The appropriate sampling plan, as outlined in the Guidelines for Sampling (CAC/GL 50-2004), Section 2.1.2 Guidelines on Sampling to control conformity of products with the specification. This should state:

- whether the specification applies to every item in a lot, or to the average in a lot, or the proportion non-conforming;
- the appropriate acceptable quality level to be used;
- the acceptance conditions of a lot controlled, in relation to the qualitative/quantitative characteristic determined on the sample.

2. Measurement Uncertainty

An allowance is to be made for the measurement uncertainty when deciding whether or not an analytical result falls within the specification. This requirement may not apply in situations when a direct health hazard is concerned, such as for food pathogens.

3. Recovery

Analytical results are to be expressed on a recovery corrected basis where appropriate and relevant, and when corrected it has to be so stated.

If a result has been corrected for recovery, the method by which the recovery was taken into account should be stated. The recovery rate is to be quoted wherever possible.

When laying down provisions for standards, it will be necessary to state whether the result obtained by a method used for analysis within conformity checks shall be expressed on an recovery-corrected basis or not.

4. Significant Figures

The units in which the results are to be expressed and the number of significant figures to be included in the reported result.

PART B: GUIDELINES ON MEASUREMENT UNCERTAINTY (CAC/GL 54-2004)

Introduction

It is important and required by ISO/IEC 17025:1999 that analysts are aware of the uncertainty associated with each analytical result and estimates that uncertainty. The measurement uncertainty may be derived by a number of procedures. Food analysis laboratories are required, for Codex purposes, to be in control[2], use collaboratively tested or validated methods when available, and verify their application before taking them into routine use. Such laboratories therefore have available to them a range of analytical data which can be used to estimate their measurement uncertainty.

These guidelines only apply to quantitative analysis.

Most quantitative analytical results take the form of "a ± 2u or a ± U" where "a" is the best estimate of the true value of the concentration of the measurand (the analytical result) and "u" is the standard uncertainty and "U" (equal to 2u) is the expanded uncertainty. The range "a ± 2u" represents a 95% level of confidence where the true value would be found. The value of "U" or "2u" is the value which is normally used and reported by analysts and is hereafter referred to as "measurement uncertainty" and may be estimated in a number of different ways.

Terminology

The international definition for Measurement Uncertainty is:

"Parameter, associated with the result of a measurement, that characterises the dispersion of the values that could reasonably be attributed to the measurand"[3]

NOTES:

1. The parameter may be, for example, a standard deviation (or a given multiple of it), or the half-width of an interval having a stated level of confidence.

2. Uncertainty of measurement comprises, in general, many components. Some of these components may be evaluated from the statistical distribution of results of a series of measurements and can be characterised by experimental standard deviations. The other components, which can also be characterised by standard deviations, are evaluated from assumed probability distributions based on experience or other information.

3. It is understood that the result of a measurement is the best estimate of the value of a measurand, and that all components of uncertainty, including those arising from systematic effects, such as components associated with corrections and reference standards, contribute to the dispersion. ."

Recommendations

1. The measurement uncertainty associated with all analytical results is to be estimated

2. The measurement uncertainty of an analytical result may be estimated by a number of procedures, notably those described by ISO (1) and EURACHEM (2). These documents recommend procedures based on a component-by-component approach, method validation data, internal quality control data and proficiency test data. The need to undertake an estimation of the measurement uncertainty using the ISO component-by-component approach is not necessary if the other forms of data are available and used to estimate the uncertainty. In many cases the overall uncertainty may be determined by an inter-laboratory (collaborative) study by a number of laboratories and a number of matrices by the IUPAC/ISO/AOAC INTERNATIONAL (3) or by the ISO 5725 Protocols (4).

[2] As outlined in Codex GL 27-1997 "Guidelines for the Assessment of the Competence of Testing Laboratories Involved in the Import and Export of Food"

[3] International vocabulary of basic and general terms in metrology, ISO 1993, 2nd Edition.

3 The measurement uncertainty and its level of confidence must, on request, be made available to the user (customer) of the results.

References

1. "Guide to the Expression of Uncertainty in Measurement", ISO, Geneva, 1993.

2. EURACHEM/CITAC Guide Quantifying Uncertainty In Analytical Measurement (Second Edition), EURACHEM Secretariat, BAM, Berlin, 2000. This is available as a free download from http://www.eurachem.ul.pt/

3. "Protocol for the Design, Conduct and Interpretation of Method Performance Studies", ed. W. Horwitz, *Pure Appl. Chem.*, 1995, 67, 33 1-343.

4. "Precision of Test Methods", Geneva, 1994, ISO 5725, Previous editions were issued in 1981 and 1986.

PART C: ISO GUIDE TO THE EXPRESSION OF UNCERTAINTY IN MEASUREMENT

In 1993 ISO published the "Guide to the Expression of Uncertainty in Measurement"[4] in collaboration with BIPM, IEC, IFCC, IUPAC and OIML. The Guide lays down general rules for the expression and evaluation of measurement uncertainty across a wide range of chemical measurements. Also included in the Guide are examples of how the concepts described in the Guide can be applied in practice. The Guide also gives an introduction to the idea of uncertainty and distinguishes between this and error, followed by a description of the steps involved in the evaluation of uncertainty.

The Guide may be applied to:

- Quality control and quality assurance in manufacturing industries
- Testing for regulatory compliance
- Testing utilising an agreed method
- Calibration of standards and equipment
- Development and certification of reference materials
- Research and development
- Both empirical and rational methods.

The Guide places emphasis on the component-by-component approach, in which the method is dissected and incremental calculations of uncertainty are made and eventually summed to provide a combined uncertainty. There has been some criticism of the practicability of this approach. Much of the work to date regarding MU has been theoretical in nature and the amount of supporting analytical data has been limited. This has caused concern to analytical chemists, especially in the food sector where analysts are already required through legislation to have some estimate of the "variability" of their results, mainly as a result of being required to use methods which have been assessed in a collaborative trial.

The evaluation of the measurement uncertainty for a method requires the analyst to look closely at all the possible sources of uncertainty within the method which may take a considerable amount of effort, although the effort involved should not be disproportionate. Usually in practice an initial study will identify the major source of uncertainty associated with the method; this will be the dominating influence on the total uncertainty of the method. It is thus possible to make a good estimate of the uncertainty for the method as a whole by concentrating on the major sources of uncertainty within the method. Following the estimation of the measurement uncertainty for a certain method in a particular laboratory, this estimate can be applied to subsequent results obtained provided that they are carried out in the same laboratory using the same method and equipment; this is of course provided that the quality control data justifies this course of action.

[4] "Guide to the Expression of Uncertainty in Measurement", ISO, Geneva, 1993.

PART D: EURACHEM GUIDE TO QUANTIFYING UNCERTAINTY IN ANALYTICAL MEASUREMENT - INTRODUCTION

EURACHEM has recently issued the second edition of its Guide to Quantifying Uncertainty in Analytical Measurement[5]. It is available as a download from www.measurementuncertainty.org.

The EURACHEM Guide is a protocol which establishes general rules for the evaluation and expression of uncertainty in quantitative chemical analysis based on the approach laid down in the ISO Guide. It is applicable at all levels of accuracy and in all fields including quality control in manufacturing, testing for regulatory compliance, calibration, certification of reference materials and research and development.

The Guide assumes that the evaluation of uncertainty requires the analyst to look closely at all the possible sources of uncertainty. It recognises that, although a detailed study of this kind may require a considerable effort, it is essential that the effort expended should not be disproportionate. It suggests that in practice a preliminary study will quickly identify the most significant sources of uncertainty, and as the examples showed, the value obtained for the total uncertainty is almost entirely controlled by the major contributions. It recommends that a good estimate can be made by concentrating effort on the largest contributions and that once evaluated for a given method applied in a particular laboratory, the uncertainty estimate obtained may be reliably applied to subsequent results obtained by the method in the same laboratory provided that this is justified by the relevant quality control data. No further effort should be necessary unless the method itself or the equipment used is changed, in which case the estimate would be reviewed as part of the normal re-validation.

In the Guide chapters 1 and 2 deal with the scope and the concept of uncertainty. Chapter 3, Analytical Measurement and Uncertainty, covers the process of method validation and conduct of experimental studies to determine method performance and their relationship to uncertainty estimation. There is also a new section on traceability. The chapter on uncertainty estimation in the previous guide has been considerably expanded and split into four separate chapters, dealing with the four steps involved in estimating uncertainty. Step 1 deals with the specification of the measurand; Step 2 with identifying the uncertainty sources; Step 3, which has been considerably expanded to cover the use of existing method validation data, deals with quantifying the uncertainty; and Step 4 covers the calculation of the combined uncertainty. The examples have been completely revised and new ones added. They are now all in a standard format, which follow the four steps described above. They all utilise the cause and effect diagram as an aid to identifying the sources of uncertainty and to ensuring that all the significant ones are included in the evaluation of the uncertainty. In addition a web site has been set up (www.measurementuncertainty.org) which contains an indexed HTML version of the Guide. This site hosts a discussion forum on the application of the guide and has a section for the publication of additional examples.

Of particular interest to food analysts are the changes since the first edition of the Guide dealing with the use of method performance data and in particular the use of method validation data, from both collaborative validation studies and from in-house studies. There are new sections dealing with the use of method performance data which show that in many cases such data gives all, or nearly all information required to evaluate the uncertainty. These new sections are of particular interest to food and feed analysts who frequently have available to them methods of analysis which are "fully validated" through collaborative trial. An important aspect is the use of cause and effect diagrams as an aid in both method validation and uncertainty evaluation. By using these diagrams it is possible to determine whether there are any components of uncertainty that are not covered by the validation data. In most cases a good validation study will provide all of the necessary data and it is possible to justify the use of an appropriate statistic, such as S_R, to determine the uncertainty.

[5] A Williams, S L R Ellison, M Roesslein (eds.), *Quantifying uncertainty in analytical measurement*, available as QUAM2000-p1.pdf., 2000, EURACHEM Secretariat, www.measurementuncertainty.org

PART E: EURACHEM GUIDE TO QUANTIFYING UNCERTAINTY IN ANALYTICAL MEASUREMENT - COMPONENT-BY-COMPONENT APPROACH

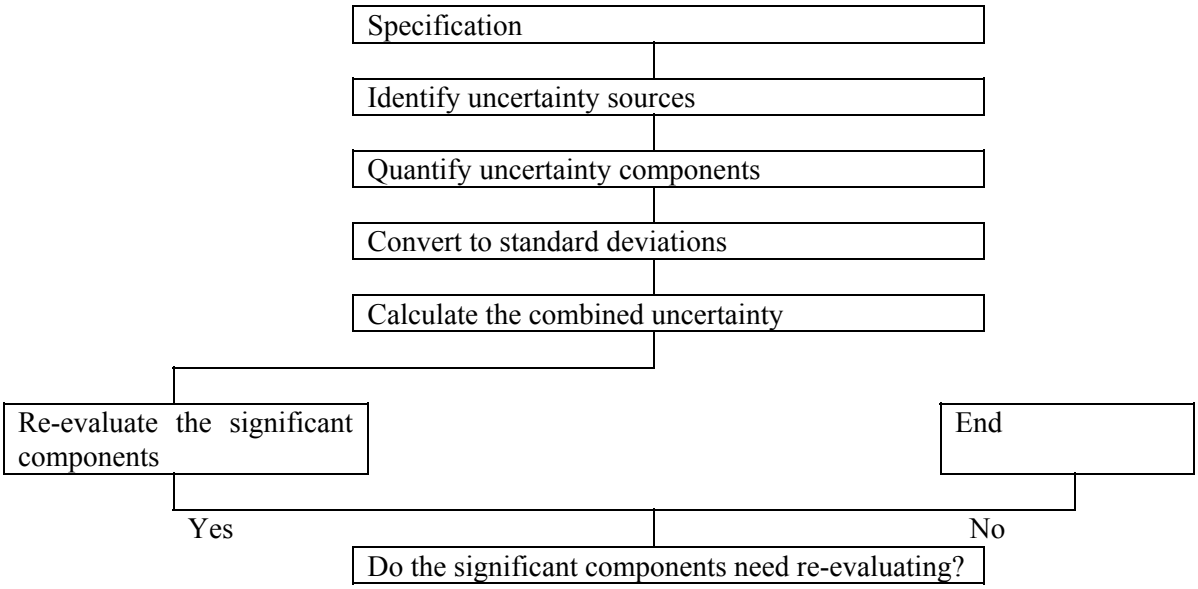
The EURACHEM Guide to Quantifying Uncertainty in Analytical Measurement is a protocol which establishes general rules for the evaluation and expression of uncertainty in quantitative chemical analysis based on the approach laid down in the ISO guide. It is applicable at all levels of accuracy and in all fields including quality control in manufacturing, testing for regulatory compliance, calibration, certification of reference materials and research and development.

Uncertainty

The word uncertainty, when used outside of the science world portrays doubt. Thus uncertainty of measurement could be understood to mean that the analyst is unsure about the validity and exactness of his result. In the EURACHEM Guide the definition attributed to uncertainty is "A parameter associated with the result of a measurement, that characterises the dispersion of the values that could reasonably be attributed to the measurand."

The Uncertainty Estimation Process

The estimation process is outlined in the EURACHEM guide and involves the steps given in the diagram below.



where:

Specification: Write down a clear statement of what is being measured and the relationship between it and the parameters on which it depends.

Identify uncertainty sources: List sources of uncertainty for each part of the process or each parameter. This is best achieved by breaking down a measurement process into a "cause-and-effect" diagram.

Quantify uncertainty components: Estimate the size of each uncertainty. At this stage, approximate values suffice; significant values can be refined in subsequent stages.

Convert to standard deviations: Express each component as a standard deviation.

Calculate the combined uncertainty: Combine the uncertainty components, either using a spreadsheet method or algebraically. Identify significant components.

The final stage is to calculate the expanded uncertainty. This is achieved by multiplying the combined standard uncertainty by a coverage factor, k. The coverage factor is chosen after considering a number of issues, such as the level of confidence required and any knowledge of underlying distributions. For most purposes a coverage factor of 2 is chosen which gives a level of confidence of approximately 95%.

Reporting Uncertainty

The information required when reporting the result of a measurement ultimately depends on the intended use but should contain enough information that the result could be re-evaluated if new data became available. A complete report should include a description of the methods used to calculate the result and its uncertainty, the values and sources of all corrections and constants used in the result calculations and uncertainty analysis and a list of all the components of uncertainty with full documentation on how each was evaluated. The data and analysis should be given in a way that it can be easily followed and if necessary repeated. Unless it is required otherwise the result should be reported together with the expanded uncertainty, U.

PART F: EURACHEM GUIDE TO QUANTIFYING UNCERTAINTY IN ANALYTICAL MEASUREMENT - USE OF COLLABORATIVE TRIAL DATA APPROACH

Section 7.6.1 of the Second Edition of the EURACHEM Guide explicitly states:

"A collaborative study carried out to validate a published method, for example according to the AOAC/IUPAC protocol or ISO 5725 Standard, is a valuable source of data to support an uncertainty estimate. The data typically include estimates of reproducibility standard deviation, s_R, for several levels of response, a linear estimate of the dependence of s_R on level of response, and may include an estimate of bias based on CRM studies. How this data can be utilised depends on the factors taken into account when the study was carried out. During the 'reconciliation' stage indicated above, it is necessary to identify any sources of uncertainty that are not covered by the collaborative study data. The sources which may need particular consideration are:

- Sampling. Collaborative studies rarely include a sampling step. If the method used in-house involves sub-sampling, or the measurand (see Specification) is estimating a bulk property from a small sample, then the effects of sampling should be investigated and their effects included.
- Pre-treatment. In most studies, samples are homogenised, and may additionally be stabilised, before distribution. It may be necessary to investigate and add the effects of the particular pre-treatment procedures applied in-house.
- Method bias. Method bias is often examined prior to or during interlaboratory study, where possible by comparison with reference methods or materials. Where the bias itself, the uncertainty in the reference values used, and the precision associated with the bias check, are all small compared to s_R, no additional allowance need be made for bias uncertainty. Otherwise, it will be necessary to make additional allowances.
- Variation in conditions: Laboratories participating in a study may tend towards the means of allowed ranges of experimental conditions, resulting in an underestimate of the range of results possible within the method definition. Where such effects have been investigated and shown to be insignificant across their full permitted range, however, no further allowance is required.
- Changes in sample matrix. The uncertainty arising from matrix compositions or levels of interferents outside the range covered by the study will need to be considered.

Each significant source of uncertainty not covered by the collaborative study data should be evaluated in the form of a standard uncertainty and combined with the reproducibility standard deviation s_R in the usual way.

For methods operating within their defined scope, when the reconciliation stage shows that all the identified sources have been included in the validation study or when the contributions from any remaining sources have been shown to be negligible, then the reproducibility standard deviation S_R, adjusted for concentration if necessary, may be used as the combined standard uncertainty."

PART G: USE OF COLLABORATIVE TRIAL: DATA – ISO 5725 CRITICAL DIFFERENCES

Note: this procedure is abstracted from the ISO 5725 Standard[6]. It was originally developed in 1981, i.e. before the concept of measurement uncertainty became formally recognised. It presumes that laboratories are operating at the same level as those which participated in the original collaborative trial to validate the method.

There is frequently available to the contractor laboratory an appropriate method of analysis which has been fully validated through a collaborative trial. The collaborative trial will give information on the analytical performance of the method, particularly the precision as expressed as the repeatability (within laboratory) and reproducibility (within and between laboratory) characteristics of the method. These values can be used to obtain a measurement uncertainty though the estimation of the so-called critical differences.

The arithmetic mean of the two single analyses obtained under repeatability conditions is compared to the (legislative or contractual) limit after calculation of the critical difference as calculated below for the analytical result.

The critical difference for the analytical result is calculated using the formula given below:

$$CrD_{95}(|\overline{Y} - \overline{m}_o|) = \frac{0.84}{\sqrt{2}}\sqrt{R^2 - r^2\,\frac{n-1}{n}}$$

where:

CrD_{95}	is the critical difference at the 95% probability value,
$\overline{Y}$	is the arithmetic mean of the results obtained,
m_o	is the (statutory/contractual etc.) limit,
n	is the number of analyses per sample,
R	is the reproducibility of the method at the concentration of interest, and
r	is the repeatability of the method at the concentration of interest.

If the difference between the (arithmetic mean) analytical result and the limit value is greater than the critical difference as calculated above, then it may be assumed that the sample which has been analysed does not fulfil the statutory or contractual requirements.

The values of r and R may have to be determined by interpolation so as to obtain the values which would apply at the limit concentration/value.

If it is to be expected that most samples comply with the statutory or contractual limit, then the final analytical results may be expected to be less than $\left[m_o + CrD_{95}(|\overline{Y} - m_o|)\right]$ if the limit is a maximum; or greater than $\left[m_o + CrD_{95}(|\overline{Y} - m_o|)\right]$ if the limit is a minimum and m_o is the given limit value.

[6] "Precision of Test Methods", Geneva, 1994, ISO 5725, Previous editions were issued in 1981 and 1986.

PART H: ISO TS 21748 - GUIDE TO THE USE OF REPEATABILITY, REPRODUCIBILITY AND TRUENESS ESTIMATES IN MEASUREMENT UNCERTAINTY ESTIMATION

INTRODUCTION

The Introduction to the ISO Guide is as given below. It demonstrates the scope of the Guide:

"Knowledge of the uncertainty associated with measurement results is essential to the interpretation of the results. Without quantitative assessments of uncertainty, it is impossible to decide whether observed differences between results reflect more than experimental variability, whether test items comply with specifications, or whether laws based on limits have been broken. Without information on uncertainty, there is a risk of mis-interpretation of results. Incorrect decisions taken on such a basis may result in unnecessary expenditure in industry, incorrect prosecution in law, or adverse health or social consequences.

Laboratories operating under ISO 17025 accreditation and related systems are accordingly required to evaluate measurement uncertainty for measurement and test results and report the uncertainty where relevant. The *Guide to the expression of uncertainty in measurement* (GUM), published by ISO, is a widely adopted standard approach. However, it applies to situations where a model of the measurement process is available. A very wide range of standard test methods is, however, subjected to collaborative study according to Part 2 of ISO 5725:1994. The present technical specification, TS 21748, provides an appropriate and economic methodology for estimating uncertainty associated with the results of these methods, which complies fully with the relevant principles of the GUM, whilst taking account of method performance data obtained by collaborative study.

The general approach used in this technical specification requires that:

- Estimates of the repeatability, reproducibility and trueness of the method in use, obtained by collaborative study as described in Part 2 of ISO 5725:1994, are available from published information about the test method in use. These provide estimates of within- and between-laboratory components of variance, together with an estimate of uncertainty associated with the trueness of the method.
- The laboratory confirms that its implementation of the test method is consistent with the established performance of the test method, by checking its own bias and precision. This confirms that the published data are applicable to the results obtained by the laboratory.
- Any influences on the measurement results that were not adequately covered by the collaborative study are identified and the variance associated with the results that could arise from these effects is quantified.

An uncertainty estimate is made by combining the relevant variance estimates in the manner prescribed by the GUM.

The dispersion of results obtained in a collaborative study may also usefully be compared with measurement uncertainty estimates obtained using GUM procedures as a test of full understanding of the method. Such comparisons will be more effective given a consistent methodology for estimating the same parameter using collaborative study data.

SCOPE OF TS 21748

The document gives guidance for

- evaluation of measurement uncertainties using data obtained from studies conducted in accordance with ISO 5725-2:1994
- comparison of collaborative study results with measurement uncertainty (MU) obtained using formal principles of uncertainty propagation (this topic is covered in section 14)

It is recognised that ISO 5725-3:1994 provides additional models for studies of intermediate precision. While the same general approach may be applied to the use of such extended models, uncertainty evaluation using these models is not incorporated in the present document.

This Technical Specification does not describe the application of repeatability data in the absence of reproducibility data.

The document is applicable in all measurement and test fields where an uncertainty associated with a result has to be determined.

The present document assumes that recognised, non-negligible systematic effects are corrected, either by applying a numerical correction as part of the method of measurement, or by investigation and removal of the cause of the effect.

The recommendations in this document are primarily for guidance. It is recognised that while the recommendations presented do form a valid approach to the evaluation of uncertainty for many purposes, other suitable approaches may also be adopted.

In general, references to measurement results, methods and processes in this document should be understood to apply also to testing results, methods and processes.

PART I: CONCEPT SET BY COMMISSION DECISION 2002/657/EC IMPLEMENTING COUNCIL DIRECTIVE 96/23/EC CONCERNING THE PERFORMANCE OF ANALYTICAL METHODS AND THE INTERPRETATION OF RESULTS

INTRODUCTION

Council Directive 96/23/EC of 29 April 1996 on measures to monitor certain substances and residues thereof in live animals and animal products[7] provides for measures to monitor substances and groups of residues listed in the Annex to the Directive. Provisions on the implementation of this Directive concerning the performance of analytical methods and the interpretation of results in this sector is given in Commission Decision 2002/657/EC of 12 August 2002 implementing Council Directive 96/23/EC concerning the performance of analytical methods and the interpretation of results[8]. This Decision provides procedures that may be followed in order to demonstrate that a specific method can be used to enforce the legislation. One of the basic concepts applied in this Directive refers to the calculation of "CCα" and "CCβ". These acronyms mean "critical concentration" characterised by a defined value for α and β using statistical terminology from hypotheses testing. This concept, when applied to the characterisation of analytical methods, is explained below.

CONCEPT

A laboratory will determine the concentration of a specific contaminant in a sample. Based on the results a decision is taken whether the measured value is:

1. above a limit indicating non-compliance or
2. below the limit.

Since the measured value has an analytical error, wrong decisions can be taken: a sample containing the analyte below the limit can wrongly be considered as non-compliant since the *measured* value is above the limit. In this Directive the Greek letter α is used to indicate the probability that this error occurs. Similarly, a sample containing the (true concentration of the) analyte *above* the limit is wrongly classified as compliant since the *measured* value is below the limit. This kind of error also has a certain probability which is described with the Greek letter β. The values CCα and CCβ indicate the concentrations at which such errors will occur with a defined probability. From the known values of CCα and CCβ laboratories may evaluate the significance of their results. Thus, in the areas of analysis to which Commission Decision 2002/657/EC applies, the values are considered important performance characteristics of a method that need to be experimentally evaluated.

Commission Decision 2002/657/EC describes various cases and different ways of calculating CCα and CCβ. In this Annex the situation in which CCα and CCβ are calculated for a method that determines a substance for which a permitted level exists is described. The situation would be different when dealing with substances *without* permitted levels.

CALCULATION

In the case that a permitted level is prescribed the measured concentration must be evaluated in order to establish whether the true but unknown concentration of the analyte is above the permitted level or not. First consider the situation in which the sample contains the analyte with a concentration just at the legislative level. Because of the analytical error (measurement uncertainty) the probability of the measured value to be below or above the limit would be equally 50 %. In consequence, all cases, in which the measured result is above the legislative level, would lead to *false positive* decisions when not taking into account the uncertainty of the result. In order to be certain that the *measured* result demonstrates that the true concentration is above the permitted or legislative limit, Commission Decision 2002/657/EC uses the CCα concept. This concentration, greater than the legislative or permitted level, is the lowest *measured*

[7] Official journal of the European Communities, L 125, 23.5.1996, p. 10
[8] Official journal of the European Communities, L 221, 17.8.2002, p. 8

concentration at which it is certain, with a given probability, that the *true* concentration is above the permitted level. Thus CCα is a *decision limit* and the risk that the true value is below the permitted limit is characterised by α. A typical value for α is 5 % indicating that the probability of a false positive result is 5 %.

EVALUATION OF CCα

The CCα concentration of a method may be established by spiking 20 blank materials with the target analyte at the permitted or legislative level and calculating the mean value of the 20 analyses along with the standard deviation of the results. CCα is equal to the mean value *plus* 1.64 times the standard deviation when accepting the probability of an α – error of 5 %.

By the development of the CCα the ability of the method to not give false positive results by focusing on samples that contain the target analyte with a concentration *below* or at the permitted level, is established.

EVALUATION OF CCβ

In contrast, when discussing CCβ, the reverse situation is considered, i.e. samples that (truly) contain the target analyte *above* the permitted level even though they analyse at below the permitted limit. This sector considers that it is important that the method clearly indicates non-compliance of the samples. However, the capability of the method to prove non-compliance depends on the true concentration of the target analyte. We look again at the example used above. Now it is assumed that the true concentration is equal to CCα. A sample containing the analyte at this concentration should be considered as non-compliant but the probability that the measured concentration is below CCα is 50 % thereby leading to *false negative* results. It may be concluded that the capability of the method to detect non-compliance is not sufficient when the true concentration is equal to the decision limit. Indeed, only if the true concentration of the sample is *above* CCα is the method capable of proving non-compliance assuming a low rate of false negative results. In particular it is of interest to establish the critical concentration at which the probability of *false negative results* for instance is below 5 % and this concentration is therefore called the detection capability, CCβ.

This value can be determined by spiking 20 blank materials with the target analyte at a concentration equal to CCα and calculating the mean value of the 20 analyses along with the standard deviation of the results. CCβ is equal to the mean value *plus* 1.64 times the standard deviation when accepting the probability of a β – error of 5 %.

COMMENT

It is important to realise that CCα and CCβ are statistically derived measures in order to limit the risk that a compliant sample is wrongly classified as non-compliant and to indicate at which concentration above the permitted limit the method is capable of proving non-compliance assuring that the rate of false negative results is sufficiently low. Moreover, these limits should not be mistaken with other performance characteristics such as limit of quantification. In fact, though CCα and CCβ are *above* the permitted level, Commission Decision 2002/657/EC also requires that the method shows sufficient trueness and repeatability at concentrations *below* the permitted level.

However, for normal enforcement analyses the CCβ concept is not used, and a concentration determined to be below the legislative limit is accepted as such in all situations.

PART J: AOAC INTERNATIONAL COMMENTS

The following paper, which has recently appeared in the *Journal of AOAC INTERNATIONAL*[9] states the AOAC INTERNATIONAL view on measurement uncertainty. It is an attempt to explain the concept of "uncertainty" as it is being widely discussed and used in the Analytical Community.

"The idea is very simple – what variability can one expect from one's measurements. But the concept was introduced initially into the analytical laboratory from metrology, which required an examination of all possible sources of error, adding them vectorially, and expanding the resulting total error statistically to arrive at a result with an attached 95% probability statement. Analytical chemists, however, had long ago realised that by performing an interlaboratory study on a standard method using a group of typical laboratories analysing a set of typical matrixes, they could reproduce almost all the uncertainty that nature could create. This practical aspect is now being incorporated into the discussion of uncertainty.

The official definition of measurement uncertainty (from the NIST Web site http://physics.nist.gov/cuu/Uncertainty/glossary.html) is:

- *Uncertainty (of measurement)*: parameter, associated with the result of a measurement that characterises the dispersion of the values that could reasonably be attributed to the measurand.

- The parameter may be, for example, a standard deviation (or a given multiple of it), or the half-width of an interval having a stated level of confidence.

- Uncertainty of measurement comprises, in general, many components. Some of these components may be evaluated from the statistical distribution of the results of a series of measurements and can be characterised by experimental standard deviations. The other components, which also can be characterised by standard deviations, are evaluated from assumed probability distributions based on experience or other information.

- It is understood that the result of the measurement is the best estimate of the value of the measurand, and that all components of uncertainty, including those arising from systematic effects, such as components associated with corrections and reference standards, contribute to the dispersion.

Considerable confusion about this term will be swept away immediately if you note that *the term "UNCERTAINTY" is attached to a RESULT, not to a method*; i.e., *measurement uncertainty* is being discussed, not *method uncertainty*. We will see how the method gets into the discussion later.

The introductory chapter to practically every textbook of quantitative analysis discusses the variability of analytical results and often advises reporting results in terms of the mean of a series of replicates and an interval within which you expect most (i.e., 95%) of your future results to fall if future analyses were conducted in an identical manner. However, the economics of chemical analysis dictates that only a few analyses are conducted on a test sample ("the results are usually good enough for government work"), so this theoretical admonition has been largely ignored until recently. Now, for accreditation purposes, laboratories are required to attach a statement of *measurement uncertainty* to their analytical results.

To obtain that halo of uncertainty surrounding your reported result, you have essentially four options:

(1) The option of calculating the equivalent of a confidence interval from the "*t*" factor applied to the standard deviation of replicates.

(2) The theoretical "bottom-up" approach recommended by the bible on uncertainty, rubber stamped by nine international organisations[10].

(3) The practical "top-down" approach from the relative standard deviation derived from an interlaboratory study by the Harmonised IUPAC/AOAC protocol[11] or ISO 5725[12].

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[9] W. Horwitz, 2003, The Certainty of Uncertainty Journal of AOAC INTERNATIONAL, **86**, 109-111

[10] "Guide to the Expression of Uncertainty in Measurement", ISO, Geneva, 1993.

[11] W. Horwitz, 1995. "Protocol for the Design, Conduct and Interpretation of Method Performance Studies", *Pure Appl. Chem.,* 1995, **67**, 331-343

(4) The estimate obtained by applying the Horwitz formula relating the relative standard deviation to concentration, as a mass fraction, $RSD_R = 2C^{(-0.15)}$, which is based upon a review of over 10,000 interlaboratory results, primarily published in the *Journal of AOAC INTERNATIONAL*.

[Alternative formula are: $\sigma_H = 0.02c^{0.8495}$ and $RSD_R = 2^{(1-0.5\log C)}$]

Option 1

Run sufficient replicates on the specific test sample under consideration to obtain a fairly good idea of how the results will scatter in routine work. If you manufacture a product to a specification of 20% fat day in and day out, with the help of a statistician, you would soon be able to know the typical uncertainty of the fat content of the product, of the sampling, and of the analysis. But if you are called upon to provide an *estimate of uncertainty* from a set of duplicates from a material you will never see again, you will have to multiply the standard deviation calculated from that pair of results by a factor of 12! Such an estimate is essentially useless because experience shows that future analysis from even a moderately experienced analyst will rarely approach the expected extreme.

Incidentally, running more replicates will not change the "true value" of the mean or of the standard deviation. More replicates provide more confidence in the interval estimate bracketing the true concentration and the true standard deviation.

Option 2

Sit down and think about everything that might possibly affect the result and estimate the expected variation that each factor will contribute to the final value. These will include uncertainties, expressed as standard deviations, from:

- standard weight corrections
- buoyancy corrections (temperature, pressure)
- volumetric flask corrections (calibration, temperature)
- pipette volume corrections (calibration, temperature)
- reference material content uncertainty
- concentration of calibrant uncertainty
- signal measurement uncertainty
- time measurement uncertainty
- extraction variability (volume, temperature, and solubility effects)
- reaction or separation variability
- effect of interferences which may or may not be present

When you have thought of everything that might possibly influence your reaction, separation, and measurement, and assigned a standard deviation to each factor, calculate the square root of the linear combination of the variances to obtain the final standard deviation that you attach to your measurement as the measurement uncertainty. Then multiply this final standard deviation by a coverage factor (k) of 2 to ensure a probability of 95%, i.e., only a 5% chance that the true value lies outside the expanded uncertainty limits. Incidentally, do not forget lot and analytical sampling, which are unique for every lot and which, therefore, require individual estimation by replication of these components for completeness. "Practical" examples can be found in a EURACHEM guide[13].

This is known as the bottom-up approach. You can come back later and add in those factors that you initially overlooked or which are pointed out to you by your colleagues or by your friendly assessor months after the report has been delivered and forgotten.

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[12] "Precision of Test Methods", Geneva, 1994, ISO 5725, Previous editions were issued in 1981 and 1986.

[13] A Williams, S L R Ellison, M Roesslein (eds.), *Quantifying uncertainty in analytical measurement*, available as QUAM2000-p1.pdf., 2000, EURACHEM Secretariat, www.measurementuncertainty.org

This absurd and budget-busting approach (for analytical chemistry) arose from metrological chemists taking over in entirety the concepts developed by metrologists for physical processes measured with 5-9 significant figures (gravitational constant, speed of light, etc.) and applying them to analytical chemistry measurements with 2 or 3 significant figures. This approach also ignores the fact that some chemical methods are influenced by numerous factors, some positive and some negative, that tend to cancel out, and that often other chemical methods are influenced by a few factors that overwhelm the weight and volume uncertainty calculations presented in the published examples.

Option 3

The approach, which is becoming generally accepted in Europe, is to conduct an interlaboratory study utilising the Harmonised IUPAC/AOAC or ISO 5725 protocol (which utilise an identical statistical model except for outlier removal). The protocols require a sample of at least 8 typical laboratories analysing a minimum set of 5 matrixes covering the range of materials of interest. Then relate the standard deviation among laboratories (S_R) as being proportional to measurement uncertainty. This is known as the top-down approach. By utilising a sample of presumably typical laboratories operating in different environments on at least 5 materials covering the range of interest, it is very likely that most of the potential error factors that are likely to be encountered in practice will have been introduced. Therefore, if we equate this S_R to measurement uncertainty and call it standard measurement uncertainty (standard uncertainty for short), we are at least about 70% certain that our result plus and minus S_R will encompass the "true" value. If we multiply S_R by a coverage factor of 2, we obtain the "expanded measurement uncertainty" (expanded uncertainty for short); we are now at least 95% certain that our result plus and minus $2S_R$ will encompass the "true" value.

When using this collaborative study approach, which results in a "standard method" as used by ISO 17025, be sure that all of the important variables are specified or understood (*see Definition of Terms and Explanatory Notes* section of the *Official Methods of Analysis of AOAC INTERNATIONAL*) with assigned limits. Weights are assumed to be within ±10% (but use the actual weight for calculations), volumetric glassware are assumed to have their assigned volume with negligible uncertainty when used with instrumental methods (but not when used in titrations), graduates are assumed to deliver the volume read from their scale, temperatures are set to be within ±2°C, pHs are within ±0.05 unit, times are followed to within 5%, and instrument scales, dials, and markers are estimated to their finest degree, then Clause 5.4.6.2 Note 2 in ISO 17025 reading, "In those cases where a well-recognised test method specifies limits to the values of the major sources of uncertainty of measurement and specify the form of presentation of the calculated results, the laboratory is considered to have satisfied this clause by following the test method and reporting instructions." Under such conditions, S_R derived from the supporting collaborative study in the same units as the reported result with the accompanying number of significant figures, usually 2 or 3, may be used as the standard uncertainty, assuming the laboratory has demonstrated that it operates within the performance limits for that method.

Option 4 or 0

As a last resort, or even before you start any analyses, you can make a rough calculation to determine if the expected uncertainty at the expected concentration will be fit for the intended purpose. Apply the Horwitz formula (or a suitably adjusted version of the Horwitz formula to account for special circumstances such as a single laboratory) to the anticipated concentration to obtain a within-laboratory S_r and multiply it by 2 to obtain the expanded uncertainty. The Horwitz formula as initially applied to among-laboratory reproducibility parameters in %, and with C expressed as a mass fraction, is

$$RSD_R\,(\text{in }\%) = 2C^{(-0.15)}$$

or as a standard deviation

$$S_R = 0.02C^{(0.85)}$$

To apply to within-laboratory repeatability parameters, divide by 2 and equate this to estimated standard uncertainty:

$$S_r = 0.01C^{(0.85)}$$

To obtain the expanded (repeatability) uncertainty, multiply by 2:

$$S_r = 0.02C^{(0.85)}$$

For example, if we are dealing with a pure compendial material, C expressed as a mass fraction is 1, so the anticipated expanded uncertainty, $2S_r$, is 0.04 or 4%. This is interpreted as 95% of anticipated results will fall between 96 and 104%. You can "improve" your uncertainty by running independent replicates. "Independent" means as a minimum "non-simultaneous" but again economics would not permit it, so the improvement would be considerably less than theoretical.

Summary: The Horwitz formula will tell you if your anticipated uncertainty is such that you will be within the limits of the ballpark with a typical method. The maximum spread obtained by the top-down approach will encompass the "true value" in almost all practical cases. It is usually easier to let nature slip in all the unanticipatable tricks that can befall even the most careful analysts than to valiantly attempt to foresee them beforehand by the budget approach. This is how the uncertainty of the method becomes entangled with the uncertainty of the measurement.

Note 1: Some of these "unanticipatable tricks" are chaotic, like dropping the thermometer or missing a decimal point. They are not subject to statistical description. Such adventitious flaws are handled by quality control but they cannot be predicted in any quantitative way. Such flaws are not intrinsic to the method.

Note 2: The uncertainty of a method, its bias and variability, is revealed by the spread of the individual measurements, i.e., by the average and standard deviation of the set of measurements. The theory envisions that an infinite set of concentration estimates is obtained for each true concentration but the hapless finite chemist is forced just to take a sampling from this infinite set at the given concentration, usually just one or two estimates. Outlier tests are applied to remove clearly extrinsic interferences with the proper application of the chemical method. Note also that the uncertainty components, both bias and variability, are functions of the true concentration, though variability is usually observed to be more concentration dependent than the bias.

If a method is to be corrected for recovery (bias) the method will usually so indicate. Many regulatory methods do not require such a correction because the specification (tolerance) was established by the same method so the recovery is "built into" the specification.

Note 3: The analytical chemist usually ignores sampling uncertainty primarily because typically little or no information accompanies the laboratory sample as to whether or not the laboratory sample truly reflects the lot. It is usually left to "management" to co-ordinate the analytical information with the sampling information. However, if the sample has been collected according to statistical principles (a process that usually requires a very large number of increments) and if these increments have been analysed to provide the basis for an estimate of sampling uncertainty, then propagation of error considerations can provide an overall "sampling + analysis" uncertainty.

Note 4: We have deliberately omitted mentioning the problem of expressing measurement and method uncertainties of microbiological examinations where the target analyte is intentionally diluted to the point of producing "true" false positives and "true" false negatives for comparison of the results from a test method to those from a reference method."

PART K: INTERNAL QUALITY CONTROL APPROACH

Laboratories which are accredited are required to have introduced acceptable internal quality control procedures. In the food sector the use of the International Harmonised Guidelines have been recommended by the Codex Alimentarius Commission.

From the use of quality control procedures it is possible to develop within laboratory estimates of repeatability and reproducibility by taking the standard deviation used within the Shewhart Charts that have been set up on introduction of the internal quality control procedures. The value here can be multiplied by 1.6 to calculate the appropriate value of reproducibility and then used as the value of σ_R in the same was as described previously.

This procedure has been used within the Netherlands Food Inspection Service (Keuringsdienst van Waren).

PART L: NMKL (NORDIC COMMITTEE ON FOOD ANALYSIS) APPROACH TO "ESTIMATION AND EXPRESSION OF MEASUREMENT UNCERTAINTY IN CHEMICAL ANALYSIS"

The first edition of the NMKL procedure was published in 1997. The procedure has been significantly revised, and re-published in 2003. It now conforms to the GUM document (Guide to the Expression of Uncertainty in Measurement, ISO, 1993), and the new EURACHEM document for measurement uncertainty.

The first edition and was a user-friendly introduction into how to estimate measurement uncertainty. It was criticised for not taking a broad enough perspective when dealing with the estimation of measurement uncertainty. This has lead to a more comprehensive approach to the subject of measurement uncertainty of quantitative analyses in the current version. Previous experience and validation data are considered when estimating measurement uncertainty, thus simplifying the estimation of the total measurement uncertainty. It is important to try to identify all sources of uncertainty in the method. A thorough review of all the steps in a method, and all sources of uncertainty, can give the analyst useful information as to where to find the major sources of error.

The procedure describes simply and clearly how to achieve good estimates of the measurement uncertainty, by using among other things, data which have been obtained from validations and other quality controls. An excel spreadsheet for calculating the combined measurement uncertainty is made available for downloading at NMKL's homepage: www.nmkl.org.

PART M. USEFUL GENERAL MEASUREMENT UNCERTAINTY REFERENCES

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