First edition
2012-01-15

Water quality — The variability of test results and the uncertainty of measurement of microbiological enumeration methods

Qualité de l'eau - Variabilité des résultats d'essais et incertitude de mesure des méthodes d'énumération microbienne

Reference number
ISO 29201:2012(E)

ISO copyright office
Case postale 56 • CH-1211 Geneva 20
Tel. + 41 22 749 01 11
Fax + 41 22 749 09 47
E-mail copyright@iso.org
Web www.iso.org

Published in Switzerland

Contents

Foreword

ISO (the International Organization for Standardization) is a worldwide federation of national standards bodies (ISO member bodies). The work of preparing International Standards is normally carried out through ISO technical committees. Each member body interested in a subject for which a technical committee has been established has the right to be represented on that committee. International organizations, governmental and non-governmental, in liaison with ISO, also take part in the work. ISO collaborates closely with the International Electrotechnical Commission (IEC) on all matters of electrotechnical standardization.

International Standards are drafted in accordance with the rules given in the ISO/IEC Directives, Part 2.

The main task of technical committees is to prepare International Standards. Draft International Standards adopted by the technical committees are circulated to the member bodies for voting. Publication as an International Standard requires approval by at least 75 % of the member bodies casting a vote.

Attention is drawn to the possibility that some of the elements of this document may be the subject of patent rights. ISO shall not be held responsible for identifying any or all such patent rights.

ISO 29201 was prepared by Technical Committee ISO/TC 147, *Water quality*, Subcommittee SC 4, *Microbiological methods*.

Introduction

Testing laboratories are required to apply procedures for estimating uncertainty of measurement (see ISO/IEC 17025[5]). Without such an indication, measurement results cannot be compared, either among themselves or with reference values (see ISO/IEC Guide 98-3:2008[7]).

General guidelines for the evaluation and expression of uncertainty in measurement have been elaborated by experts in physical and chemical metrology, and published by ISO and IEC in ISO/IEC Guide 98-3:2008.[7] However, ISO/IEC Guide 98-3:2008[7] does not address measurements in which the observed values are counts.

The emphasis in ISO/IEC Guide 98-3:2008[7] is on the “law of propagation of uncertainty” principle, whereby combined estimates of the uncertainty of the final result are built up from separate components evaluated by whatever means are practical. This principle is referred to as the “component approach” in this International Standard. It is also known as the “bottom-up” or “step-by-step” approach.

It has been suggested that the factors that influence the uncertainty of microbiological enumerations are not well enough understood for the application of the component approach (see ISO/TS 19036:2006[6]). It is possible that this approach underestimates the uncertainty because some significant uncertainty contributions are missed. Reference [19] shows, however, that the concepts of ISO/IEC Guide 98-3:2008[7] are adaptable and applicable to count data as well.

Another principle, a “black-box” approach known as the “top-down” or “global” approach, is based on statistical analysis of series of repeated observations of the final result (see ISO/TS 19036:2006[6]). In the global approach it is not necessary to quantify or even know exactly what the causes of uncertainty in the black box are.

According to the global philosophy, once evaluated for a given method applied in a particular laboratory, the uncertainty estimate 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 (EURACHEM/CITAC CG 4[10]). Every analytical result produced by a given method thus should have the same predictable uncertainty. This statement is understandable against its background of chemical analysis. In chemical analyses the uncertainty of the analytical procedure and the uncertainty of the final result of analysis are usually the same. The global principle dismisses the possibility that there might be something unique about the uncertainty of a particular analysis.

The uncontrollable “variation without a cause” that always accompanies counts alters the situation for microbiological enumerations. The full uncertainty of a test result can be estimated only after the final result has been secured. This applies to both the global and the component approaches.

The unpredictable variation that accompanies counts increases rapidly when counts get low. The original global design is therefore not suitable for low counts, and therefore also not applicable to most probable number (MPN) methods and other low-count applications, such as confirmed counts.

It is often necessary, and always useful, to distinguish between two precision parameters: the uncertainty of the technical measuring procedure (operational variability), which is more or less predictable, and the unpredictable variation that is due to the distribution of particles. A modification of the global principle that takes into account these two sources of uncertainty is free from the low-count restriction. This is the global model detailed in this International Standard.

In theory, the two quantitative approaches to uncertainty should give the same result. A choice of two approaches is presented in this International Standard. Offering two approaches is appropriate not only because some parties might prefer one approach to the other. Depending on circumstances one approach may be more efficient or more practical than the other.

Neither of the main strategies is, however, able to produce unequivocal estimates of uncertainty. Something always has to be taken for granted without the possibility of checking its validity in a given situation. The estimate of uncertainty is based on prior empirical results (experimental standard uncertainties) and/or reasonable general assumptions.