INTERNATIONAL STANDARD

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In vitro diagnostic medical devices — Measurement of quantities in samples of biological origin — Description of reference materials

Dispositifs médicaux de diagnostic in vitro — Mesure des grandeurs dans les échantillons d'origine biologique — Description des matériaux de référence

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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 3.

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 International Standard may be the subject of patent rights. ISO shall not be held responsible for identifying any or all such patent rights.

International Standard ISO 15194 was prepared by the European Committee for Standardization (as EN 12287:1998) and was adopted, under a special “fast-track procedure”, by Technical Committee ISO/TC 212, *Clinical laboratory testing and* in vitro *diagnostic test systems*, in parallel with its approval by the ISO member bodies.

Contents

Foreword

This European Standard has been prepared by Technical Committee CEN/TC 140 "In vitro diagnostic medical devices", the secretariat of which is held by DIN. International Federation of Clinical Chemistry (IFCC) and the European Confederation of Laboratory Medicine (ECLM) have contributed to its preparation.

This European Standard shall be given the status of a national standard, either by publication of an identical text or by endorsement, at the latest by December 1999, and conflicting national standards shall be withdrawn at the latest by December 1999.

This European Standard has been prepared under a mandate given to CEN by the European Commission and the European Free Trade Association, and supports essential requirements of EU Directive(s).

For relationship with EU Directive(s), see informative Annex ZA, which is an integral part of this standard.

This European Standard is based on ISO Guide 31 "Contents of certificates of reference materials". The future European Standard "Measurement of quantities in samples of biological origin – Presentation of reference measurement procedures" presents requirements to ensure that values assigned to reference materials by such procedures are reliable and stated in a useful way.

Annexes A, B and ZA are for information only.

According to the CEN/CENELEC Internal Regulations, the national standards organizations of the following countries are bound to implement this European Standard: Austria, Belgium, Czech Republic, Denmark, Finland, France, Germany, Greece, Iceland, Ireland, Italy, Luxembourg, Netherlands, Norway, Portugal, Spain, Sweden, Switzerland and the United Kingdom.

Introduction

To produce useful and reliable results of measurement, whether in science, technology, or routine service, it is necessary that they are supported by a reference measurement system so as to be comparable and ultimately traceable to measurement standards of the highest metrological level.

The substances which are used to obtain this traceability, both through time, distances, and different measurement procedures, are the reference materials. A given reference material is supported by documentation containing descriptions, measurement results, instructions for use, stability data, and storage conditions. The present European Standard specifies the content and format of such supporting documentation.

Reference materials are used for one of three main purposes:

a) calibration of values indicated by a measuring system or of another reference material;

b) validation or control of trueness of measured values in a given laboratory, or in a group of laboratories;

c) evaluation of the performance of a new measurement procedure.

The maximum acceptable uncertainty of measurement of the assigned value of the reference material depends on the requirements of the results of the measurement procedure.

As the proper use of a reference material depends on its description, it is important to apply rules for the documentation of reference materials.

The advantages of having (written) standards available are listed in ISO/IEC Guide 15.

1 Scope

This European Standard specifies requirements and formats for the description of reference materials. It is applicable to reference materials of higher metrological order, classifiable as primary measurement standards and secondary measurement standards that function either as calibrators or control materials for reference measurement procedures. This standard does not apply to reference materials that are parts of an in vitro diagnostic measuring system.

This European Standard also provides instructions on how to collect basic data for value determination and how to present the assigned value. The standard also specifies the format for a certificate.

This European Standard is not applicable to the production of the reference materials.

2 Normative references

This European Standard incorporates by dated or undated reference, provisions from other publications. These normative references are cited at the appropriate places in the text and the publications are listed hereafter. For dated references, subsequent amendments to or revisions of any of these publications apply to this European Standard only when incorporated in it by amendment or revision. For undated references the latest edition of the publications referred to applies.

EN 375 : 1992, *In vitro diagnostic systems – Requirements for labelling of in vitro diagnostic reagents for professional use.*

ISO 31 : 1992, *Quantities and units.*

3 Terms and definitions

For the purposes of this Standard, the terms and definitions given in International Vocabulary of Basic and General Terms in Metrology apply (3.1 and 3.2 are quotes from VIM) together with the following:

3.1
primary measurement standard
standard that is designated or widely acknowledged as having the highest metrological qualities and whose value is accepted without reference to other standards of the same quantity [6.4 of VIM]

NOTE 1: The concept of primary standard is equally valid for base quantities and derived quantities.

NOTE 2: The word "measurement" has been included in the term here for consistency.

NOTE 3: Measurement standards include reference materials.

3.2
secondary measurement standard
standard whose value is assigned by comparison with a primary standard of the same quantity [6.5 of VIM]

NOTE 1: The word "measurement" has been included in the term here for consistency.

NOTE 2: Measurement standards include reference materials.

3.3
matrix (of a material system)
all components of a material system except the analyte

3.4
matrix effect
influence of a property of the sample, independent of the presence of the analyte, on the measurement and thereby on the value of the measurable quantity

NOTE 1: A specified cause of a matrix effect is an influence quantity.

NOTE 2: A matrix effect depends on the detailed steps of the measurement as described in the measurement procedure.

EXAMPLE:
The measurement of the amount-of-substance concentration of sodium ion in plasma by flame emission spectrometry may be influenced by the viscosity of the sample.

3.5
commutability of a material
ability of a material to yield the same numerical relationships between results of measurements by a given set of measurement procedures, purporting to measure the same quantity, as those between the expectations of the relationships obtained when the same procedures are applied to other relevant types of material

NOTE: For reference materials used to calibrate measurement procedures intended for biological samples, "other relevant types of material" include a large number of samples from healthy and relevantly diseased individuals.

3.6
report
document giving detailed information on a reference material, supplementary to that contained in a certificate or package insert

NOTE: The information may comprise the preparation of the reference material, methods of measurement, factors affecting trueness, statistical treatment of results, and the way in which traceability was established.

4 Classification and naming of reference materials

4.1 Description of the properties of reference materials

A reference material has properties, each of which shall be described according to the following format:

a) system (i. e. the material itself);

b) any relevant component(s); and

c) kind-of-quantity (quantity in a general sense).

If the property is a measurable quantity, it shall have a value that is equal to

d) a numerical value multiplied by

e) a unit of measurement.

EXAMPLE:
Certified reference material(BCR; CRM 303)--Calcium(II); amount-of-substance concentration (reconstituted) c = 2,472 mmol/l (U = 0,019 mmol/l; k = 2) where U is the expanded uncertainty of measurement using the coverage factor k

4.2 System

4.2.1 Function

The reference material shall be capable of functioning as one of the following:

a) calibration material (calibrator): to determine the calibration function of a given measurement procedure (which may then also be used to calibrate another reference material); or

b) control material: to assess the analytical trueness or uncertainty of measurement of an established or new measurement procedure in a given laboratory or in a group of laboratories.

Within a given measuring system in a given laboratory, a reference material shall perform only one of the functions, and which one shall be specified by the term calibration material (calibrator) or control material.

4.2.2 Application and authorities

The application and authorities of the system shall consist of the following:

a) geographical scope; e. g. international, regional, national, local;

b) responsible body, e. g. authority, institution, company, or laboratory;

c) certification of properties where applicable.

NOTE: Examples of the responsible bodies of item b) include the World Health Organization (WHO), the Standards, Measurements and Testing Programme of the European Commission, national reference institution, manufacturer of separately available reference materials or a specialized hospital laboratory. In item c) some values may have to be certified, e. g. by the Standards, Measurements and Testing Programme of the European Commission or by the National Institute of Standards and Technology (NIST) of USA. Other values may be given for information only.

4.2.3 Hierarchical position of reference materials of higher order in the reference measurement system

Measurement standards of higher order shall be classified according to their positions in the reference measurement system for a given quantity as follows:

a) primary measurement standard (see 3.1);

b) secondary measurement standard (see 3.2).

4.2.4 Identification code(s)

An identification code shall be assigned to the reference material. A lot identification shall be included when available.

4.2.5 Reference material characteristics

The characteristics of the reference material shall be described in the following terms:

a) The origin and nature of the starting material: inorganic, organic, synthetic, natural, or biological of a stated species, e. g. human, porcine.

b) The matrix:

– simple, when a reference material consists of a pure component in a well-defined medium;

– complex, when the component is present in a partially known milieu, e. g. stabilized blood or serum.

c) The physical state of the analyte in the material: solution, colloid, or suspension.

d) The physical state of the reference material: solid, liquid, or gas.

e) Homogeneity and phases: a description of the reference material in terms of one or more individual homogeneous or inhomogeneous phases, e. g. blood can be considered a combination of a homogeneous plasma phase and an inhomogeneous cell phase.

4.3 Component(s)

The component(s) shall be named according to an internationally accepted nomenclature, including for example any necessary indications of elementary entity, relative molecular mass or molar mass, oxidation state, multiple forms comprised, for enzymes the EC number.

EXAMPLES:
Aliphatic carboxylate (C_{10} to C_{26}, non-esterified), fibrinogen (340 000), iron (II+III), and lactate dehydrogenase (E.C.1.1.1.27) isoenzyme 1, basic fibroblast growth factor (human, rec. DNA).

4.4 Kind-of-quantity (quantity in a general sense)

The kind-of-quantity (quantity in a general sense), e. g. mass, amount-of-substance, number fraction, substance concentration, shall always be stated. If no simple relationship between component and system can be expressed, reference shall be made to the measurement procedure.

NOTE 1: "Quantity in a general sense" is the VIM term for what IFCC/IUPAC has called kind-of-quantity.

NOTE 2: Appropriate names and symbols for kind-of-quantities (quantities in the general sense) are given in ISO 31 and in publications by IFCC and IUPAC.

4.5 Numerical value

The number of significant figures of the result shall be chosen so that the uncertainty of measurement lies on the last or – if the first significant figure of the uncertainty measure is 1 or 2 – on the two last figures. For numerical values with more than four figures on either side of the decimal mark they should be separated by a space in groups of three counting from the mark to the left or right.

4.6 Unit of measurement

The unit chosen shall be an SI unit or other legal unit whenever possible.

NOTE: WHO defines some off-system units called International Units.

4.7 Construction of systematic designations and trivial names

A systematic name and value shall consist of elements as specified in 4.2 to 4.6 and shall be given in the format as specified in 4.1.

EXAMPLE 1:
Primary reference material for calibration/control(WHO IS XXX; certified; lyophilized)--Component; kind-of-quantity = (xxx ± xx) unit; average and expanded uncertainty giving an interval estimated to have a level of confidence of 0,95.

EXAMPLE 2:
A systematic name of a calibrator for a haematology analyser can be

Secondary reference material for calibration (Responsible body NN; Product no YYYY)--

Erythrocytes; number concentration = (xxx ± vv) unit; average and expanded uncertainty (level of confidence of 0,95)

Leukocytes; number concentration = (vvv ± zz) unit; average and expanded uncertainty (level of confidence of 0,95)

Thrombocytes; number concentration = (zzz ± yy) unit; average and expanded uncertainty (level of confidence of 0,95).

4.8 Trivial names

A trivial name shall be constructed by omitting from the systematic name elements that are not necessary for the understanding of the function of the reference material in the measurement.

EXAMPLE 1:
The trivial name for the material given in Example 1 of 4.7 can be WHO(IS XXX)--Component.

EXAMPLE 2:
The trivial name in general form for the material given in Example 2 of 4.7 can be

Calibrator(Responsible body NN; Product no YYYY)--Erythrocytes, Leukocytes and Thrombocytes;

or even

Calibrator(Responsible body NN; Product no YYYY)--Blood cells.

The trivial name for the corresponding individual product can be

Calibrator(Company NN; Product no YYYY; Batch no AAAA)--Blood cells.

5 Description of a reference material

5.1 Elements of a description

The description of a reference material of higher metrological order shall comprise at least the elements listed as mandatory (M) in Table 1.

NOTE: The order of the elements listed in Table 1 may be changed and additional elements, such as an abstract, may be added as appropriate.

Table 1: Main elements (clauses) of a report describing a reference material of higher metrological order

Element	Type [1])		Subclause in this European Standard
	M	O	
Title page	I		
Contents list		I	
Foreword	I		
Warning and safety precautions	N		5.2
Introduction		I	5.3
Title of report	N		
Scope	N		5.4
Definitions		N	
Symbols and abbreviations		N	
Terminology		N	5.5
Justification for choice of reference material	I		5.6
General characteristics	I		5.7
Specific characteristics	I		5.8
Validation	I		5.9
Intended function	I		5.10
Instructions for use	I		5.11
Supplier	I		5.12
Bibliography		I	5.13
Annexes		I	5.14
Dates	I		5.15

[1]) Symbols for type of element in a European Standard:
M mandatory, O optional;
I informative, N normative.

5.2 Warning and safety precautions

5.2.1 Attention shall be drawn to any danger associated with the reference material and its use. All necessary precautions shall be described. Regional, national, and local legislation and regulations may apply.

5.2.2 This information shall be printed in capital letters or in bold type as follows:

a) immediately after the title of the standard if the danger encountered is due to the reference material, e. g. native material of human origin with, in principle, potential infectiveness (but found negative for HIV antibody, hepatitis B virus surface antigen, and hepatitis C virus antibody), radioactive material, or a carcinogen;

b) as a cautionary statement under instructions for use, e. g. for a measurement using equilibration gases (CAVE aerosol formation).

Warning notes and safety precautions shall be unnumbered.

NOTE: The source text presenting the dangers to health should be quoted where appropriate.

5.3 Introduction

The introduction shall comprise the following items, as appropriate, in any order:

a) names of measurable quantities in the measurement of which the reference material is intended to be used, indicated by system, component, and kind-of-quantity (quantity in a general sense);

b) systematic description of the reference material according to clause 4;

c) main reasons for the selection of the particular base material for the reference material and for the chosen method of stabilization (e. g. bovine haemolysate stabilized by freezing);

d) reference measurement procedure(s) or other measurement procedure(s) used for assigning values to the reference material;

e) statement that the requirements of a reference material have been met:

– traceability to measurement procedures or reference materials of higher metrological order (when such exist);

– organization of any collaborative studies and the summary results obtained from material-certification studies;

– number of laboratories participating in any such studies;

– number fraction of results collected during the study that were rejected according to stated rules.

5.4 Scope

The clause shall define the subject and aspect(s) covered, indicating limits of applicability.

NOTE: The items include as appropriate

a) current reference measurement procedure(s) or current generally-used routine methods of measurement or measurement procedures for which the reference material is produced;

b) methods of measurement or measurement procedures for which the material is known to be unsuitable;

c) influence quantities in the reference material involving, e. g. drugs, metabolites, additives, microbial growth;

d) mentioning of major required pretreatment of the reference material which is not performed on the biological samples according to a specified measurement procedure (for example, reconstitution of lyophilized material).

5.5 Terminology

5.5.1 General

This element shall describe the meaning and use of concepts and terms that are specific to the description, unfamiliar to potential readers, or chosen among several possibilities for a stated reason.

NOTE: The clause "Terminology" is complementary to the clause "Definitions" (see table 1) and sometimes also to the clause "Symbols and abbreviations" (see table 1), and the terms may be incorporated in either or distributed between both.

5.5.2 Nomenclature

The names of measurable quantities as well as their spelling and structure shall be taken from the latest recommendations by authoritative sources.

The names of kind-of-quantities (quantities in a general sense), their symbols and units shall be in accordance with the European or International Standards, especially the ISO 31 series.

5.5.3 Trivial names

If a trivial name is used it shall be given in parentheses following the systematic name the first time the systematic name appears in the text.

5.6 Justification for choice

The production and selection of reference materials is a multifactorial optimization and should be governed by clinical needs, production possibilities, metrological needs, analytical problems, economic, ethical and safety considerations, and the requirements needed for the end-use. Such considerations shall be stated.

5.7 General characteristics

5.7.1 The origin of the starting material shall be stated in accordance with the terms given in 4.2.5.

5.7.2 The relevant historical details of the starting material shall be stated so far as they influence the properties of the final material, e. g. age and sex of donors, temperature and length of storage of blood serum and clot together before separation, storage time and temperature. The safety aspects shall be covered, e. g. the testing of material of human origin for hepatitis B virus surface antigen, hepatitis C virus antibody, HIV antibody, and other markers of infectiveness as stipulated by regulations on each donated portion.

5.7.3 Details of sample preparation of the starting material shall be described. Purification with a check for impurities shall be indicated.

5.7.4 The compounds and concentrations of any additives shall be specified.

EXAMPLE:
Additives for reference materials used in the clinical laboratory include anticoagulants, antioxidants, antimicrobial agents, stabilizers, wetting agents, and coating of pellets.

5.7.5 The physical state and phases of the reference material shall be stated in accordance with the terms given in 4.2.5.

5.7.6 The estimated within- and between-sample homogeneity, with regard to the minimum analytical portion, shall be stated (see 5.9.2).

EXAMPLE:
A certified reference material consists of a lyophilized tissue powder filled into vials. The within-vial homogeneity was investigated on three replicate samples, each having a mass of 200 mg, taken from the material in each of 20 vials. The between-vial variation was investigated on one sample of 200 mg from each of 60 vials selected from various positions on the lyophilization plate. The instructions for use states that "the recommended minimum analytical portion has a mass of 200 mg".

5.7.7 If applicable, the physical form shall be described, e. g. shape, dimensions, number, and amount.

EXAMPLE:
A certified reference material, glass filters for light spectrometry, is intended as a reference source for the calibration of the absorbance scale of spectrometers. It consists of three individual filters and one empty filter holder, 10 mm × 10 mm × 50 mm. Each filter bears an identification number. The upper left corner has been removed to indicate correct placing in the metal holder.

5.7.8 Any sterilization procedure applied shall be described.

5.7.9 The container and/or packaging shall be specified as to type, material, closure, and atmosphere.

5.7.10 Storage conditions for the unopened container shall be given, e. g. temperature, humidity, and light. The extent of instability under the prescribed conditions shall be stated. The frequency with which the stability of the material is to be checked during the life of the lot shall be stated (see 5.9.4). It shall be stated if the reference material is of restricted stability once its container is opened.

NOTE: Such reference materials are usually supplied in sealed units.

5.7.11 The quality systems which were followed during production, handling, storage, and distribution shall be identified, e. g. according to the ISO 9000 series.

5.7.12 Any hazard associated with the reference material or its use shall be stated and appropriate precautions detailed (see 5.2).

5.8 Specific characteristics

5.8.1 The specific characteristics of a reference material that influence any quantity for which a value is given shall be described by at least the information specified in 5.8.2 to 5.8.12.

5.8.2 When available the molecular composition or biological function of each relevant component shall be stated in accordance with 4.3.

5.8.3 The quantity to which a value is assigned shall be specified.

5.8.4 The purity of the main component in a "pure" reference material shall be stated as mass fraction, volume fraction, amount-of-substance fraction, or number fraction. The unchanged biological activity of a potentially unstable biological component (analyte) shall be stated.

5.8.5 The matrix of the material shall be described in accordance with 4.2.5 b).

NOTE: Important information for dried and lyophilized materials is the proportion of any solvent residue, and this should be stated when relevant.

5.8.6 The measurable quantity for which a value is given shall be adequately stated with indications of system, component, and kind-of-quantity, and with relevant specifications to each (see 4.1).

5.8.7 Data for the extent of commutability investigated shall be given where applicable, e. g. for catalytic concentration of enzymes.

5.8.8 The type of scale on which the value of the quantity is measured shall be specified, that is whether it is a nominal, ordinal, difference (also called interval), or ratio scale.

NOTE: The set of possible values should be given if required.

EXAMPLE:
A stabilized blood sample may be used as a reference material for the measurement of the amount-of-substance concentration difference of base-binding groups ("base excess") in blood and a difference scale is required (.... -4,2 -4,1 ... -0,1 0,0 0,1 ... 4,1 4,2 ....) mmol/l; the amount-of-substance concentration of hydrogen carbonate ion is measured on a ratio scale (0,0 0,1 0,2 ....) mmol/l.

5.8.9 SI units shall be used wherever possible and appropriate. Where an arbitrary unit is used it shall have an internationally agreed definition or a definition described by a given measurement procedure.

5.8.10 The uncertainty of measurement shall be expressed either as a combined standard uncertainty or as an interval derived from the expanded uncertainty with a stated level of confidence.

NOTE: The components of the uncertainty of measurement are caused by inhomogeneity of the material and – during the value-assigning exercise – by its instability and by analytical variation among laboratories, operators, calibrations of measuring systems, measurement procedures, and runs. They comprise systematic as well as random components.

EXAMPLE 1:
Certified reference serum(no. 348 NN, reconstituted)--Progesterone; amount-of-substance concentration c = (40,3 ± 1,0) nmol/l; unweighted average and expanded uncertainty giving an interval estimated to have a level of confidence of 0,95.

EXAMPLE 2:
The certified purity of a certain material is stated as relative amount-of-substance content (actual/theoretical) = 0,9963 (0,9936; 0,9975); median (0,25- and 0,75-fractile).

5.8.11 The traceability of assigned values of reference materials shall be described.

5.8.12 The quality assurance implemented in the laboratories providing values for the reference material shall be referenced.

5.9 Validation

5.9.1 Planning of experimental design

The material certification study shall be described.

NOTE: The study will depend on the nature of the reference material and the way in which it will be used. Generally its homogeneity and stability should be investigated before considering the assignment of values. It is also necessary to decide on the maximum permissible uncertainty of measurement of the assigned value, because this will influence the design of the study.

5.9.2 Assessment of homogeneity

Studies on the homogeneity of the reference material within-sample and between-sample shall be described.

NOTE: Supplementary details, including statistical treatment, are given in ISO/IEC Guide 35.

5.9.3 Statistical evaluation of results

A statistical evaluation of the data collected during the study shall be made. The method of evaluation shall be described.

NOTE: The data should be scrutinized for consistency and outliers in accordance with ISO 5725-2 and further evaluated in accordance with ISO/IEC Guide 35, or alternatively non-parametric approaches may be used.

5.9.4 Assessing the stability

The procedures for assessing the stability shall be described, taking appropriate regional, national, and local legislation and regulations into consideration, and the results presented with appropriate statistics.

NOTE 1: For reference materials of higher order, a stability allowing storage life from 8 to 10 years of use is a relevant aim. It is often possible to estimate how the decomposition will take place with time by exposing samples for relatively short periods (e. g. several weeks) to a range of temperatures, including some at which deterioration will occur more rapidly than at the proposed storage temperature.

The effect of relevant influence quantities on stability shall be documented.

The measurement procedures shall be described, including their calibration and accuracy control.

NOTE 2: Procedures for monitoring the stability of the reference material during its lifetime involve measurement of characteristic quantities at planned intervals during the time in which the reference material will be used; for instance measurement of haemoglobin concentration in the plasma of a reference preparation of stabilized blood.

EXAMPLE:
Vials of the materials were stored at -20, 37, 45, and 56 °C and measured in duplicate after storage for 110, 244, and 604 days. No statistically significant changes were found in the respective values relative to the values of samples stored at -20 °C at which temperature the material had been shown to be stable when compared to a sample stored at -90 °C. The material seems sufficiently stable. The stability will be checked during the lifetime of the material.

5.9.5 Value assignment

The experimental plan and the reference measurement procedures used in assigning values shall be described (see also ISO/IEC Guide 35).

NOTE: The value for a given quantity may be assigned on the basis of one very well documented measurement procedure in one laboratory (see 5.9.6). In many cases, however, a better foundation is obtained when several experienced laboratories are involved in a collaborative study at a high metrological level and, if possible, using different methods of measurement or even different principles of measurement.

5.9.6 Value and uncertainty of measurement assigned by one measurement procedure in one laboratory

The reference measurement procedure of the highest metrological level shall be selected. Details, including an uncertainty budget, shall be described or referred to so that it can be reproduced in other laboratories.

NOTE 1: A log describing actually performed adjustment and maintenance of equipment, and reports on validation of the measurement procedure and control data should be kept.

The following experimental elements shall be reported:

a) number of replicates within-run;

b) number of runs;

c) time interval between each run;

d) number of different operators;

e) number of calibrations;

f) number of different measuring systems for the same purpose;

g) suppliers and lots of reagents.

The final result(s) and certified value(s) shall be accompanied by their respective uncertainties (see 5.8.10).

NOTE 2: Supplementary details are given in ISO/IEC Guide 35 and ISO 5725-2.

5.9.7 Regional recognition

Any regional recognition of the reference material shall be listed.

5.10 Intended function

The intended function of the reference material shall be stated (see 4.2.1). Known limitations of suitability shall be documented, e. g. when a calibrator or control material intended for use with a particular field measurement procedure gives measurement values that require a correction different from that applied to values on native material. The recommended statistical treatment of the values obtained by the user shall be indicated for each of the intended functions, at least by reference to literature.

EXAMPLE 1:
The uncertainty of measurement of a calibration may be calculated from the uncertainty of calibration provided with the assigned value of the calibration material and the repeatability standard deviation of the measurement procedure, taking into account the number of measurements obtained on the calibrator. Formulae for calculation are presented in ISO/IEC Guide 33.

EXAMPLE 2:
In order to check the precision of a measurement procedure performed in a laboratory, the number of replicate measurements on the reference material is selected according to the acceptable probability of erroneous acceptance or false rejection of the run. The assessment of trueness is made by comparing the average value of observed data with the assigned value taking into account the uncertainties of measurement of both. Elimination of outliers may be needed. Formulae for calculation are presented in ISO/IEC Guide 33.

EXAMPLE 3:
When using the reference material for control of trueness in each analytical run including four determinations on the reference material, one control rule may be: no observation shall exceed the $\pm 3s_r$ limits of acceptance. In this way a systematic error of $2s_r$ is detectable with a probability of 0,55 and the probability of false rejection is 0,01. (The repeatability standard deviation of the measurement procedure is symbolized by s_r.)

5.11 Instructions for use

5.11.1 Safety

The first paragraph shall contain any cautionary statement (see 5.2). Safety precautions concerning equipment, materials, samples, and waste shall be included where appropriate in the instructions for use.

5.11.2 General

Detailed instructions for use shall be provided, including at least the following information as appropriate:

a) required storage conditions and stability of the reference material as received and after opening of the outer container;

b) opening of the immediate container;

c) preparation of sample;

d) techniques for achieving thawing or reconstitution followed by mixing;

e) procedure for obtaining the minimum analytical sample and analytical portion;

f) measurement procedure (either recommended or mandatory);

g) required storage conditions and stability of the reference material after opening the immediate container;

h) disposal of any remaining material after use.

5.11.3 Reagents

If reagents are cited in the instructions for use, each item shall be identified.

5.11.4 Apparatus

Apparatus required for use of a reference material shall be listed.

5.11.5 Environment

If the use of the reference material requires specific environmental conditions, these shall be stated.

5.11.6 Measuring volumes

When necessary, the temperature (and for gases, the pressure) at which a volume is measured shall be specified.

Dilutions prepared by adding one volume of liquid to a volume of another shall be indicated by

a) "diluted $V_1 \rightarrow V_2$" if the volume V_1 of the specified solution is diluted in such a way as to give a total volume V_2 of final mixture, e. g. diluted 25 ml $\rightarrow$ 1 l; or

b) "diluted $V_1 + V_2$" if the volume V_1 of the specified solution is added to the volume V_2 of the solvent, e. g. 25 ml + 975 ml.

Expressions such as "V_1:V_2" or "V_1/V_2" shall not be employed as they are used with different meanings.

5.11.7 Reconstitution of lyophilized reference material

Details of reconstitution shall be given.

NOTE: Lyophilized reference materials are the most frequent type of reference material in the clinical laboratory.

5.11.8 Reference to patented items

If, in exceptional cases, technical reasons justify the preparation of a European Standard in terms which include the use of a patented item, the rules given in Annex A of ISO/IEC Directives – Part 2: 1992 shall apply.

5.12 Supplier

The identity of the supplier of the reference material shall be stated.

5.13 Bibliography

Documents which contain additional information, but which are not necessary in order to use the reference material for its intended function, shall be listed in a bibliography.

NOTE 1: This bibliography may take the form of an annex (see 5.14). It may include documents which:

a) are not publicly available;

b) only serve for information;

c) have merely served as references in the preparation of the standard.

NOTE 2: The types of publication may comprise, e. g., regional and national standards, industrial standards, legal regulations, recommendations issued by international and regional scientific organizations, scientific journal papers, textbooks, manufacturers' standard or product literature, and interlaboratory trial reports.

5.14 Annexes

Data and information that do not fit into the main part of the report shall be given in annexes.

NOTE: Such items may include

a) data on homogeneity (see 5.9.2);

b) data on value assignment (see 5.9.5, 5.9.6);

c) bibliography (see 5.13);

d) labels (see clause 6);

e) certificate (see clause 7);

f) package insert (see clause 8).

5.15 Dates of authorization and revision

The dates for the current publication and of any previous edition(s) shall be given.

6 Label

The labels of the immediate container and the outer container should be in accordance with EN 375. The product name on the label shall be in accordance with clause 4.

7 Certificate

Certificate shall include the items specified in ISO Guide 31 and, in addition, the following as appropriate:

a) identification of the individual portion of the reference material;

b) sources or origin of the reference material;

c) method of preparation of the reference material;

d) transportation;

e) instruction for the correct handling of the reference material, including any storage conditions, shelf-life, and stability after a seal is broken;

f) certified value and uncertainty for each measurable quantity with number of accepted values obtained during the trial;

g) some indication of statistical treatment;

NOTE 1: Basic data obtained by individual laboratories and methods are usually referred to an annex or to a report.

h) if appropriate, non-certified values ("recommended values", "indicative values") and values obtained by procedures of lower metrological levels, all provided with estimates of measures of uncertainties;

i) use of values for calibration and for control of trueness (often with reference to the full report);

j) methods of measurement used for the certification;

k) any planned checks of stability;

l) names of operator, responsible scientist, and/or laboratories participating in various parts of the trial;

m) reference to a larger report containing all relevant information;

n) supplier of the reference material if different from the certifying body;

o) annexes as specified above. The use of annexes is also relevant to achieve a reasonably small size of the main text of a certificate;

p) date of certification.

NOTE 2: If there is a certificate, it should be issued by a recognized metrological body.

All information on the origin, preparation, and any additions to the reference material shall be given, i. e. no information is omitted due to confidentiality.