INTERNATIONAL FEDERATION OF CLINICAL CHEMISTRY (IFCC)¹, ²

Scientific Division

Expert Panel on Theory of Reference Values³ (EPTRV)

Approved Recommendation on the Theory of Reference Values

Part 4. Control of Analytical Variation in the Production, Transfer and Application of Reference Values

Prepared for publication ⁴ by

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Preface

This paper is the fourth in a series of Recommendations on the Theory of Reference Values. Other parts deal with:

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Part 1. The Concept of Reference Values (1).

Part 2. Selection of Individuals for the Production of Reference Values (2).

Part 3. Preparation of Individuals and Collection of Specimens for the Production of Reference Values (3).


Part 6. Presentation of Observed Values Related to Reference Values (5).

A Guide to the Documents is also in preparation.


⁴) Received for publication 1991-07-08.
The Expert Panel on Theory of Reference Values (EPTRV) was created in 1970 by the Committee on Standards (later changed to Scientific Committee and then to Scientific Division) of the International Federation of Clinical Chemistry (IFCC). Its task was to develop a nomenclature and recommend procedures for the production of reference values and their treatment, and presentation of observed values in relation to reference data.

The first document in the above-mentioned series describes the subject of reference values and defines various terms. It should be consulted prior to the reading of the present document for a thorough understanding.

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2. Monitoring procedures
3. Transfer of reference value data
4. Comment (to Schemes 1 and 2)
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1. Introduction

1.1. Aim of the document

Observed values may be compared with reference values or with the reference distribution, reference limits and reference interval derived from the reference values (1, 5).

The purpose of this document is to provide guidance on the requirements necessary to ensure that the reference values are produced using accurate and precise methods. Failure to employ methods that have known and suitably low levels of inaccuracy and imprecision reduce clinical utility of the values obtained.

1.2. Perspective

It is important to establish and maintain a realistic awareness of the contribution that analytical inaccuracy and imprecision may have on the production of reference values. The biological variance is an important factor to take into consideration when determining the allowable imprecision and inaccuracy (6—8). Thus, every effort should be made to establish the level of analytical inaccuracy and imprecision tolerable for the intended uses of the reference values. For example, the Aspen Conference defined in 1976 the tolerable level of imprecision for measurements of calcium and urea in serum to be quite different, 0.9 per cent and 6.2 per cent (CV), respectively (cited in l.c. (6)).

This document assumes that adequate effort has been made to reduce biological variation in selecting subjects (2) and collecting specimens (3).

1.3. Need for reliable methods

The method, including the reagents and equipment used, must be described in enough detail that adequately-trained analysts will all proceed in the same manner and obtain reproducible results. The assessment of these methods should be made according to Part 2 of the Recommendations of the IFCC's Expert Panel on Nomenclature and Principles of Quality Control (9).

1.4. Need for long-term stability of the analytical method

The development of reference values is a time-consuming and costly exercise. As a result, it is essential that the values must have application over a lengthy period. Furthermore, the true clinical utility of the values in diagnosis, comparative assessment and long-term monitoring necessitates the employment of well controlled analytical procedures.

1.5. Experimental design

In the production of reference values it is desirable to keep analytical variance of approximately the same magnitude as that obtained by good-quality routine analysis. Therefore, it is strongly recommended that the measurements for the production of reference values be made under strictly controlled, but realistic routine conditions, including the day-to-day imprecision.

1.6. Transferability

The complexity, cost and effort of establishing reference values are often minor compared to the problem of obtaining a sufficient number of adequate specimens for the production of the reference values. As a result it is often necessary to transfer reference values from one institution to another. In this case there is an essential need for laboratories involved to obtain comparable results. That can be achieved by assessment of the accuracy and precision of the analytical method in use through long-term inter-laboratory studies (10).

Comparability of results is also necessary for laboratories receiving specimens from a highly mobile population. It may be best achieved using similar analytical procedures. Should they be different, it is important that they conform to tolerable limits of inaccuracy and imprecision.
2. Monitoring Procedures

2.1. Theoretical Principles Underlying Quality Control

The theoretical principles underlying quality control (QC) and the various forms QC can take are treated in detail in the recommendation of the Expert Panel on Nomenclature and Principles of Quality Control in Clinical Chemistry (9).

There are numerous QC procedures which will monitor the accuracy and precision of the analyses, and ensure the transferability of the data. This section provides elementary guidance in QC procedures for this purpose. It is assumed that laboratories performing this work will have certain materials and facilities available which will, probably, be in use routinely. However, it is essential that the following conditions are met:

- The calibration material must be carefully defined (9, 11), keeping in mind the presence of non-specific components which may contribute to the reading.

- The basis for calculation, e.g., molar lineic absorbance (9, 11), must be carefully defined and tested.

- Suitable control specimens must be available commercially or be prepared by the user. They should have a matrix that has properties similar to those of the specimens used for the production of reference values (7–11). To ensure proper monitoring of accuracy, precision and reliability of a method, selected control materials should be validated (10).

- The control procedure should be described in detail. This description should also include decision rules for out-of-control situations and actions to perform when an alarm occurs (12).

2.2. Recommendation for Single Laboratory Scheme

In most cases, a simple system involving control specimens is recommended for this purpose because of its general applicability (9). The recommended system (13, 14) consists of internal and external quality control procedures, see Scheme 1. (External quality control is also called external quality assessment.)

The procedures for the two separate kinds of internal control, control of precision and control of accuracy, are shown in Scheme 2.

<table>
<thead>
<tr>
<th>Scheme 1. Basic program for quality control</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Internal quality control</strong></td>
</tr>
<tr>
<td>- Control of precision</td>
</tr>
<tr>
<td>- in every run of analyses, even if the run consists of only one patient specimen</td>
</tr>
<tr>
<td>- with precision control specimen</td>
</tr>
<tr>
<td>- Control of inaccuracy</td>
</tr>
<tr>
<td>- in at least every 4th run</td>
</tr>
<tr>
<td>- with accuracy control specimen</td>
</tr>
<tr>
<td><strong>External quality control (or assessment)</strong></td>
</tr>
<tr>
<td>- Interlaboratory surveys</td>
</tr>
<tr>
<td>- at least three times a year for each type of quantity</td>
</tr>
<tr>
<td>- specimens shipped by the chief investigator</td>
</tr>
<tr>
<td>- evaluation on the basis on analytical results from independent reference laboratories</td>
</tr>
</tbody>
</table>

In accuracy control it is important that the whole clinically relevant interval of measurement be monitored.

In some cases control rules are needed that are more sensitive to inaccuracy and imprecision than those described in Schemes 1 and 2. For example, it is important to detect small increases in inaccuracy and imprecision of the analysis of the concentration of calcium in serum because of the small biological variance of this component. Westgard, Groth and co-workers have developed control systems that may ensure that the analytical quality is within stated limits (12, 15).

2.3. Recommendation for Multiple Laboratory Scheme

If several laboratories are to participate in the production of reference values, the comparability of the results from these laboratories must first be evaluated. If comparability is inadequate, appropriate changes must be made in the analytical system. Whenever several laboratories are involved, there must be ongoing checks on comparability while reference values are being produced.

Prior to analyzing the specimens from the reference individuals the laboratories should assess the comparability of their analytical results by using the same control material. Where stability of the component permits, it is advisable also to check comparability by exchanging native human specimens between laboratories. An effective model for conducting and evaluating such a long-term survey has been developed and experience with the model has been reported (10). If the data from the comparison study show that no significant effect on the size of the reference interval will be the result, then a multi-laboratory study can proceed.
Scheme 2: Recommended basic internal quality control

<table>
<thead>
<tr>
<th></th>
<th>Control of precision</th>
<th>Control of accuracy</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Frequency</strong></td>
<td>In every run of analyses</td>
<td>In every 4th run of analyses</td>
</tr>
<tr>
<td><strong>Materials</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Control specimens</td>
<td>A precision control specimen from a batch used for as long a time as possible</td>
<td>One of a number of different accuracy control specimens kept on hand</td>
</tr>
<tr>
<td>Concentration at the decision limit</td>
<td>Assigned values at normal and pathological levels</td>
<td></td>
</tr>
<tr>
<td>Monitoring device</td>
<td>Control chart</td>
<td>Form for assessment of accuracy</td>
</tr>
<tr>
<td><strong>Task of analyst</strong></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Recognizes control specimen</td>
<td>Recognizes control specimen</td>
<td></td>
</tr>
<tr>
<td>Knows concentration</td>
<td>Does not know concentration (known to control supervisor of the laboratory)</td>
<td></td>
</tr>
<tr>
<td>Conducts duplicate determinations</td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Goals</strong></td>
<td>Assessment of random variation (random errors)</td>
<td>Detection of systematic deviations (systematic errors)</td>
</tr>
<tr>
<td></td>
<td>Detection of trends</td>
<td>Monitoring over the whole diagnostically relevant interval</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Detection of influence of other components (matrix)</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Elimination of intentional or unintentional misinterpretation of data by analyst</td>
</tr>
</tbody>
</table>

3. Transfer of Reference Value Data

The diversity of analytical procedures has complicated the establishment of transferable reference values. Therefore, employment of the above mentioned QC procedures with well operated and documented methodology is a prerequisite for the transfer of reference value data from one laboratory to another; so is also the comparability of the populations involved. However, it is important to realize that the clinical environment is dynamic, and that reference values and associated data may become invalid or inappropriate over a period of time. To prevent the obsolescence of reference value data it is imperative to ensure the following:

- Monitor changes in method accuracy and precision.
- Check relevance of reference value data to individuals and/or groups being assessed.
- Store the original reference value data.
- Update reference value information when necessary.

5. Appendix: Methods of Quality Controls

A simple procedure for the control of accuracy and precision is shown in Schemes 1 and 2.

5.1. Accuracy control

It is essential to detect changes in the calibration curve over time and to monitor the specificity of the analytical method.

This requires a number of accuracy control specimens at both normal and pathological levels of concentrations of the type of quantity for which reference values are to be produced. These control specimens should contain quite different kinds of matrix, and should also contain other components that might disturb the analysis. Accuracy and specificity are not monitored adequately if the different concentrations of the target component are obtained by spiking the same matrix or by adding different amounts of diluent to the same amount of the specimen during reconstruction.

5.2. Precision control

To ensure that the required precision is maintained and to detect trends, it is recommended that an adequate number of control specimens be included at fixed or random positions in each analytical run (7, 8). Ideally precision control should be employed at different levels of concentration. If only one precision control is used the concentration should coincide with the clinical discrimination level.
6. References


13. Regulations and explanations regarding the implementation of the guidelines of the Medical Society of West Germany (1974) Mitteilungen der Deutschen Gesellschaft für Klinische Chemie 5, 33—43; Deutsches Ärzteblatt 71, 961—965.
