

EPo6-A

Evaluation of the Linearity of Quantitative Measurement Procedures: A Statistical Approach; Approved Guideline

This document provides guidance for characterizing the linearity of a method during a method evaluation; for checking linearity as part of routine quality assurance; and for determining and stating a manufacturer's claim for linear range.

A guideline for global application developed through the Clinical and Laboratory Standards Institute consensus process.

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Evaluation of the Linearity of Quantitative Measurement Procedures: A Statistical Approach; Approved Guideline

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Abstract

CLSI document EP06-A—*Evaluation of the Linearity of Quantitative Measurement Procedures: A Statistical Approach; Approved Guideline* is intended to provide both manufacturers and users of quantitative analytical methods with an economical and user-friendly method of establishing or verifying the linear range. This guideline also can be used to demonstrate the extent to which a quantitative analytical method meets clinical requirements or manufacturer's linear range claims.

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SAMPLE

Evaluation of the Linearity of Quantitative Measurement Procedures: A Statistical Approach; Approved Guideline

1 Scope

This document presents a method to establish, verify and/or demonstrate the linear range of a quantitative measurement procedure. These methods do not identify the causes of significant nonlinearity. The method employs increasing numbers of samples for more definitive examinations of linearity. Therefore, if a failed demonstration is evaluated and it is determined that the experiment needs to be repeated, it can be done with more replicates or with fewer levels to cover a smaller range.

- This protocol is to assess linearity, isolated as much as possible from conditions of precision and trueness. It is understood that poor precision will hinder an effective assessment of linearity, so a check for poor repeatability is included.
- These experiments should use samples with a matrix appropriate to the specimens being analyzed (serum, plasma, urine, etc.)
- This protocol requires laboratories to set goals for nonlinear error. It provides basic concepts for setting such goals, but does not recommend any specific protocol.

2 Introduction

2.1 Purpose

The purpose of this guideline is to describe a statistical process for determining the linearity of a quantitative measurement procedure. This primary objective is to determine the concentration(s) where a method is not linear and the extent of the nonlinearity at that level. This guideline emphasizes the necessity that each user establishes his or her requirements for linearity, or the allowable error due to nonlinearity. It also places less importance on global tests for linearity across the tested range (such as the LoF test). Global tests merely indicate that statistically significant nonlinearity exists; they do not show where that nonlinearity is, nor do they show the magnitude of the error. Linearity tests can be helpful to assess bias, which is a component of measurement error, but nonlinearity is not the only component of bias.

Users should have an understanding of their needs for measurement error, bias, random error (or imprecision), and nonspecificity (or interferences). From these they can derive a goal for linearity. In this context, NCCLS document EP06-A is a part of a series of documents that guide users through the process of method evaluation. Also see the most recent version of NCCLS document EP21—*Total Analytical Error for Clinical Laboratory Methods*.

This document is meant to cover a broad range of situations, such as establishing the linear range of a method, which requires testing across a wide range of concentrations, then progressively narrowing into a range of acceptable linearity. It is also intended to cover situations where the linear range has been determined elsewhere (e.g., by the manufacturer), but the user wishes to verify that range in their laboratory. The same procedure is used for all scenarios, but with different numbers of concentration levels and different numbers of replicates.

2.2 Alternative Approaches

There are many experimental approaches used to evaluate claims of a linear range. Kroll and Emancipator^{1,2,3} proposed a method based on comparison of a linear regression equation to a series of polynomial equations, using the residuals (distances from each data point to the calculated regression line) to define linearity. The advantages of this method are that it is robust and based on rigorous scientific principles. Demonstration that the linear (first-order) fit is the best provides evidence that the method is linear.

The process is not simple, because users must be able to test a variety of polynomial equations against the experimental data (although spreadsheet programs can be used). These procedures were further refined by Kroll, Praestgard, Michaliszyn, and Styer⁴ and implemented in the College of American Pathologists interlaboratory comparison programs. Krouwer and Schlain⁵ proposed a general method using the “last point off the line” (LPO) method, which requires iterative calculations of a multiple least-squares equation. Tholen⁶ proposed a graphic evaluation along with a regression lack of fit compared to a goal percentage; and alternatively a simple analysis of the slopes of consecutive line segments.

2.3 Definitions

Allowable difference/Allowable error - The magnitude of analytical deviation, from all sources, that a user can tolerate in a testing system and still meet the medical requirements of the test; **NOTE:** The allowable difference (error) boundaries (for a single observation) are represented by the target value of the specimen plus or minus the allowable error amount.

Analyte - Component indicated in the name of a measurable quantity (*ISO FDIS 17511*).⁷

Analytical result - For this document, the final result of a measurement on a test specimen; **NOTE:** The result is usually in concentration or activity units; it is assumed that the measurement procedures to be evaluated by the procedures in this guideline are quantitative methods that yield a numerical output.

Bias - The difference between the expectation of the test results and an accepted reference value;⁸ **NOTE:** In general, the deviation/difference is based on replicate measurement using an accepted (definitive, reference, or designated comparison) method and the method being tested, and expressed in the units of the measurement or as a percentage.

Input variable - The given value that is used as a reference (independent variable) and against which the output variable is compared; **NOTE:** The input variable is represented by X with individual values of X noted by x_i (i represents an individual observation); its value is plotted along the X-axis (abscissa).

Intercept//Y intercept - **1)** The point where a function intersects an axis; **2)** The value of a variable, when the value for the other variables is zero; **NOTE:** The y-intercept is the value of the y-variable when the x-variable has a value of zero.

Least squares regression - The method of statistically placing the location of the estimated line or curve among the data so that the sum of the squares of the distances of each data point from the line in the perpendicular direction from the X-axis (i.e., parallel to the Y-axis) is minimized; **NOTE:** It allows the direct algebraic computation of the coefficients and an estimate of their uncertainty.

Linear equation - An equation that represents a straight line; **NOTE:** A linear equation is typically represented mathematically by: $Y = a + bX$; X and Y are the input and output variables, b is the slope, and a is the y-intercept.

The Quality System Approach

NCCLS subscribes to a quality system approach in the development of standards and guidelines, which facilitates project management; defines a document structure via a template; and provides a process to identify needed documents through a gap analysis. The approach is based on the model presented in the most current edition of NCCLS HS1—*A Quality System Model for Health Care*. The quality system approach applies a core set of “quality system essentials (QSEs),” basic to any organization, to all operations in any healthcare service’s path of workflow. The QSEs provide the framework for delivery of any type of product or service, serving as a manager’s guide. The quality system essentials (QSEs) are:

- | | | | |
|---------------------|------------------------|------------------------|------------------------|
| Documents & Records | Equipment | Information Management | Process Improvement |
| Organization | Purchasing & Inventory | Occurrence Management | Service & Satisfaction |
| Personnel | Process Control | Assessment | Facilities & Safety |

EP06-A addresses the quality system essentials (QSEs) indicated by an "X." For a description of the other NCCLS documents listed in the grid, please refer to the Related NCCLS Publications section on the next page.

Documents & Records	Organization	Personnel	Equipment	Purchasing & Inventory	Process Control	Information Management	Occurrence Management	Assessment	Process Improvement	Service & Satisfaction	Facilities & Safety
					EP9 EP10 EP14 EP15 EP21				EP7		M29

Adapted from NCCLS document HS1—*A Quality System Model for Health Care*.

Related NCCLS Publications*

- EP5-A** **Evaluation of Precision Performance of Clinical Chemistry Devices; Approved Guideline (1999).** This document provides guidance for designing an experiment to evaluate the precision performance of clinical chemistry devices; recommendations for comparing the resulting precision estimates with manufacturers' precision performance claims and determining when such comparisons are valid; as well as manufacturer's guidelines for establishing claims.
- EP7-A** **Interference Testing in Clinical Chemistry; Approved Guideline (2002).** This document provides background information, guidance and experimental procedures for investigating, identifying, and characterizing the effects of interfering substances on Clinical Chemistry test results.
- EP9-A2** **Method Comparison and Bias Estimation Using Patient Samples; Approved Guideline – Second Edition (2002).** This document addresses procedures for determining the bias between two clinical methods or devices, and for the design of a method comparison experiment using split patient samples and data analysis.
- EP10-A2** **Preliminary Evaluation of Quantitative Clinical Laboratory Methods; Approved Guideline—Second Edition (2002).** This guideline provides experimental design and data analysis for preliminary evaluation of the performance of an analytical method or device.
- EP14-A** **Evaluation of Matrix Effects; Approved Guideline (2001).** This document provides guidance for evaluating the error or bias in analyte measurements that is due to the sample matrix (physiological or artificial) when two analytical methods are compared.
- EP15-A** **User Demonstration of Performance for Precision and Accuracy; Approved Guideline (2001).** This document describes the demonstration of method precision and accuracy for laboratory analyte determinations utilizing a protocol designed to be completed within five working days or less.
- EP21-A** **Estimation of Total Analytical Error for Clinical Laboratory Methods; Approved Guideline (2003).** This document provides manufacturers and end users with a means to estimate total analytical error for an assay. A data collection protocol and an analysis method which can be used to judge the clinical acceptability of new methods using patient specimens are included. These tools can also monitor an assay's total analytical error by using quality control samples.
- M29-A2** **Protection of Laboratory Workers from Occupationally Acquired Infections; Approved Guideline—Second Edition (2001).** Based on U.S. regulation, this document provides guidance on the risk of transmission of hepatitis viruses and human immunodeficiency viruses in any laboratory setting; specific precautions for preventing the laboratory transmission of blood-borne infection from laboratory instruments and materials; and recommendations for the management of blood-borne exposure.

* Proposed- and tentative-level documents are being advanced through the NCCLS consensus process; therefore, readers should refer to the most recent editions.



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