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Laboratory Statistics—Standard Deviation; A Report

Clinical and Laboratory Standards Institute ™

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Providing NCCLS standards and guidelines, ISO/TC 212 standards, and ISO/TC 76 standards

This report provides two methods for calculating standard deviation, as well as rules for significant figures and rounding of results. Sample data sets are included for evaluating computer-generated statistics.

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Laboratory Statistics—Standard Deviation; A Report

ABSTRACT

Laboratory Statistics — Standard Deviation; A Report (NCCLS document EP13-R) is written for laboratorians as well as manufacturers. It provides the correct method for calculating standard deviation and the means to test software. Procedures for determining the standard deviation by both the one-pass and two-pass procedures, as well as the rules for determination of significant figures and rounding of results, are described. Specific sample data sets are included for use when employing computer-generated statistics to verify that the program is working correctly. References from the statistical and clinical laboratory literature concerning many relevant statistics are provided.

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FOREWORD

This document provides the correct method for calculation of standard deviation and the means to test (validate) related software. It describes procedures for determining the statistic standard deviation, as well as the rules for determination of significant figures and rounding of results. Two different methods for calculation of standard deviation, (1) the one-pass procedure and (2) the two-pass procedure, are described. Specific sample data sets are included to be used when employing computer-generated statistics to provide standard results that have been peer reviewed. The user will thus have model data and results so that if identical results are generated by the computer program in question, the user will know that the program is working correctly. Finally, this document lists references from the statistical and clinical laboratory literature concerning many relevant statistics.

The Area Committee for Evaluation Protocols reviewed this document and recommended that it be issued as a report, rather than as a "guideline" or "protocol," because it presents basic technical information. A report is a document that has not been subjected to consensus review and is released by the NCCLS Board of Directors.

Key Words

Coefficient of variation, degrees of freedom, Gaussian curve, population, rounding, sample, significant figures, standard deviation, variance.

Laboratory Statistics — Standard Deviation; A Report

1.0 INTRODUCTION

The ability to produce reliable statistical calculations for mean, standard deviation, and variance is a requirement of good laboratory practice. A key aim within the clinical laboratory is to minimize errors in order to give the best possible results for a patient. The variability of results is measured by a descriptive term called the standard deviation. It represents a special point on the general Normal curve where data distribution begins to spread out.

This document provides the following items for the laboratorian and manufacturer:

- The definitions and formulas for mean, standard deviation, and variance

- A method to check the accuracy of standard deviation computations. The document contains sample problems with solutions for the statistics of standard deviation and variance.

- Rules and procedures for the generation of a computed answer with respect to the number of significant figures and the correct procedures for rounding the data.

2.0 STANDARD DEVIATION

2.1 Symbols Used in Text

Definitions of the terms mean, standard deviation, and variance are expressed differently in text books and spread sheets. For example, the following symbols have been used to designate standard deviation: s, sd, SD, and STD. Similarly, the symbols s^2, var, and VAR represent the term variance. In this document, the following symbols are used:

	Population	**Sample**
standard deviation	σ	s
variance	σ^2	s^2
mean (average)	μ	$\bar{x}$
number of observations	N (n)	N (n)
degrees of freedom	N (n)	N−1 (n−1)

2.2 Population versus Sample

When an entire population of data is summarized by a few statistics, these statistical terms are called population parameters. Population parameters are known, or can be calculated exactly, since one knows about all the items in the population. However, if only some portion of the total population of items or data have been observed, then one can say that a sample of the total population has been observed. One can infer what the total population of items or data look like from the sample of the data. This sampling of items or data can be summarized with sample statistics. A sample standard deviation is an estimate of the population standard deviation.

Clinical laboratory scientists use statistics to describe properties of data. For example, one might analyze a serum sample 20 to 30 times for glucose. The resulting data will have a distribution of results from a low value to a high value. Usually, this distribution of results is symmetrical with what is known as a Gaussian distribution. The more precise the analysis, the narrower the distribution of data points; the less precise the analysis, the wider the distribution of data points.

Specimens received in the laboratory do not represent the entire population with respect to a specific analyte. Following are two cases that illustrate the difference between population parameters and sample statistics:

Population parameter: A marble collector has 5 bags of marbles. There are a total of 50 marbles, although each bag may contain a variable number of marbles. In this case, the total number of marbles in the collection can be counted and is exact. One obtains the population mean (average) of each bag by dividing 50 by 5.

Sample statistics: Five bags of marbles are taken from an assembly line at a marble factory for quality control purposes. In this case, the number of marbles in each bag on the assembly line can only be estimated by this sample of 5 bags. In the table below, the population statistics and the sample statistics are compared for these two examples of 50 marbles with 5 bags.

Bag Number	Number of Marbles in Bag
1	10
2	9
3	10
4	11
5	10

	Population Parameter		Sample Statistics	
mean	10	(μ)	10.0	($\bar{x}$)
variance	0.40	(σ^2)	0.50	(s^2)
standard deviation	0.632	(σ)	0.707	(s)

Because the number of statistical calculations performed in the clinical laboratory is large, laboratory scientists depend on the personal computer to do their calculations. The manufacturer of a leading spreadsheet program started providing a formula to compute only the population standard deviation instead of sample standard deviation statistics. Other software manufacturers followed suit. Personal computers use commercial and noncommercial software to do calculations. Also, manufacturers of many clinical biochemistry instruments incorporate software to calculate these statistics.

2.3 The Two Types of Standard Deviation and Formulas for Calculation

Two types of standard deviation, one for sample statistics and one for population parameters, have been described. There are two ways to perform the calculations associated with each of these, a manual method (two-pass) and a computer method (one-pass). In theory, both methods will give the same mathematical result; however, in practice results can differ, depending on how many decimal places (positions) are used in the calculations. The formulation of the manual method can be expanded and recognized in a different algebraic way to yield the formulation for the computer method.

2.3.1 *Manual Method (Two-Pass Procedure)*

The manual method is used to calculate the mean for each list of values. Subtract the mean value from each of the values and square the difference. Add together the sum of the squares to get a sum of the squares of the differences.

(1) If the sum of the squares of the differences is divided by the number of data values (N or n), the value is termed the population variance. The square root of the population variance will give the population standard deviation.

(2) If the sum of the squares of the differences is divided by one less than the number of data values (N−1 or n−1), the value is termed the sample variance. The square root of the sample variance will give the sample standard deviation.

$$\sigma = \sqrt{\frac{\sum_{i=1}^{i=n} (x_i - \mu)^2}{n}} = \text{population standard deviation}$$

$$s = \sqrt{\frac{\sum_{i=1}^{i=n} (x_i - \bar{x})^2}{n-1}} = \text{sample standard deviation}$$

2.3.2 *Computer Method (One-Pass Procedure)*

The computer method is used to calculate three values from the data values. These calculated values are (1) N or n, the number of data values, (2) a cumulative sum of the "x" values, and (3) a cumulative sum of the "x" values squared. With these three values, one can calculate the mean, variance, and standard deviation values. If one uses only the N (n) value in the denominator, then one will derive the population value for the variance. The square root of this number will give the population standard deviation. If one uses only the value N−1 or n−1 in the denominator, then one will derive the sample variance. The square root of this number will give the sample standard deviation.

$$\sigma = \sqrt{\frac{\sum_{i=1}^{i=n} x_i^2 - \frac{\left(\sum_{i=1}^{i=n} x_i\right)^2}{n}}{n}} = \text{population standard deviation (computers)}$$

$$s = \sqrt{\frac{\sum_{i=1}^{i=n} x_i^2 - \frac{\left(\sum_{i=1}^{i=n} x_i\right)^2}{n}}{n-1}} = \text{sample standard deviation (computers)}$$

2.3.3 *Coefficient of Variation*

The coefficient of variation (CV) is a statistical measurement of the relative variation (dispersion) of data points of a sample based on a 100% scale. The formulation for CV, which is expressed in a percentage, is:

$$CV = \frac{S * 100}{\bar{x}}$$

2.3.4 *Properties of the Standard Deviation*

The standard deviation is a measure of reproducibility, the variation of repeated measurements of the same sample. But there are many other statistics that measure reproducibility. The range (the largest minus the smallest observation) is one example. The standard deviation (or the CV) is widely used for the following reasons:

One advantage of the standard deviation is its statistical properties:

- The expected value of the standard deviation does not change if one changes the sample size, assuming that the original sample size was reasonable. Multiple determinations of the sample standard deviation will cluster around the same value (the population standard deviation, σ), so one does not have to adjust for different sample sizes between experiments.

- The sample standard deviation (s) has less sampling error than other statistics that might be done to determine variability of the results. For example, if one were to repeat a reproducibility experiment 10 times, the 10 standard deviations would have less variation than the 10 ranges calculated from the same data.

Another advantage of the standard deviation is that it can be used with the mean to completely describe the Gaussian distribution (see Figure 1):

- Most repeated measurements in the clinical laboratory will form a symmetric, bell-shaped distribution around a central value. The mathematical function that describes the distribution of measurements is the Gaussian (or Normal) density function.

- The Gaussian density function has two parameters: the mean (μ) and standard deviation (σ). These are estimated by the sample mean ($\bar{x}$) and the sample standard deviation (s).

- One can use the properties of the Gaussian distribution to make detailed statements about the reproducibility of the measurements. For any Gaussian distribution, the following are true:

$$\mu \pm 1\sigma = 68.27\%;$$

$$\mu \pm 2\sigma = 95.45\%;$$

$$\mu \pm 3\sigma = 99.73\%.$$

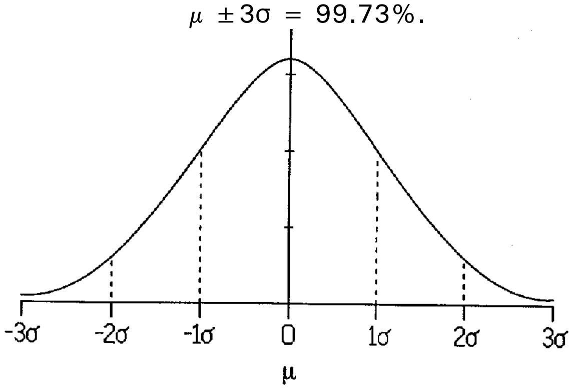

Figure 1. Gaussian (Normal) curve: area under curve.

2.4 Correct Use of Degrees of Freedom

When an entire population is examined, it is mathematically permissable to determine the value for the standard deviation using a formula with the value for the number of observations (N) in the denominator. When patient results for biochemical tests are used, one has only a small sample from the whole population and the value of the number of observations minus 1 (N-1) in the denominator to determine the value for standard deviation. The term for this value (N−1) is called the degrees of freedom and it is used in the calculation of the sample variance to give an unbiased estimate of the population variance.

When using a value of N in the denominator, the value obtained is called the standard deviation of the population. When using the value of N−1 in the denominator, the value obtained is called the standard deviation of the sample. The latter value, N−1, should be used in determining the standard deviation when sampling in the laboratory.

The laboratory data are samples of a general population, and thus, one can infer from the sampling a description of the entire population of results or samples. It is always samples of the general population that are being measured and from this one can infer a description of the entire population. It has been found that when the value N−1 is used in the denominator, this is a best estimate of the standard deviation of the general population.[5,6] See Appendix A for examples that use N−1.

The statistical mathematical functions for s and s^2 that have been utilized in many spreadsheet programs may not be correct. This can be particularly true with older versions of the software. Some newer versions have been changed to include two formulations for both standard deviation and variance. The statistical value for variance is the square of the standard deviation value. The difference in calculation using N versus N−1 becomes less significant as N increases.

2.5 Computational Methods for Standard Deviation

Both the one-pass and two-pass systems will generate the same answers in the absence of a rounding error. The use of the one-pass system is preferred because the error will never be greater than that with the two-pass system. If one rounds (truncates) intermediate results during the calculation process, significant errors can occur.

RECOMMENDATION: Use a one-pass system with the maximum precision available; do not round intermediate results.

3.0 REPORTING OF RESULTS

3.1 Rounding of Data

3.1.1 *Manual Method*

Rounding of data has been described in standard texts on biostatistics.[1,2,3,5] The basic rules for rounding are as follows:

1. The right-most digit to be rounded is not changed if it is followed by a digit less than 5.

   Examples: 3.464 is rounded to 3.46

   1.44 is rounded to 1.4

2. If the right-most digit to be rounded is followed by a digit greater than 5, or by 5 followed by other non-zero digits, it is increased by one.

   Examples: 2.7351 is rounded to 2.74

   1.7456 is rounded to 1.75

3. The odd/even rule: When the right-most digit to be rounded is odd and it is followed by a 5, which is followed by zeros, then the digit is increased by one (similar to rule #2). If the right-most digit to be rounded is even and it is followed by a 5, which is also followed by zero digits, then the even number is unchanged.

   Examples: 2.7350 is rounded to 2.74 (note 3 is odd)
   1.7450 is rounded to 1.74 (note 4 is even)

3.1.2 *Automated Method*

The automated approach used by all computer and calculator software is different in the sense that all digits, regardless of whether they are even or odd, are increased by one when they are followed by 5 or greater.

Examples: 2.7350 is rounded to 2.74
1.7450 is rounded to 1.75

3.2 Significant Figures

There have been many rules proposed for determining the number of significant figures (digits) in an answer.[5] Most of these deal with significant figures (digits) in individual measurements. For the purposes of this report, a distinction is made between significant figures in patient reportable results and significant figures in method evaluation.

Significant figures in reportable results may be based on the significant figures in the calibrator and the resolution power of the measurement system, or some other information, such as medical needs. For evaluating methods, modify the format of reportable results as follows:

- When calculating the mean, use the number of decimal digits in the reportable result plus one.

- For standard deviation calculations, use the number of decimal digits plus one, or two significant figures, whichever has more decimal digits.

- When calculating coefficient of variation, use two significant figures or one decimal digit, whichever is greater.

If insufficient significant figures are used in the evaluation of methods, significant errors in estimating the statistical parameters can result.

4.0 PRECAUTIONS FOR COMPUTER USERS

Just because a computer program is prepared by a well-known organization, do not assume that it cannot be flawed. <u>Laboratory scientists are responsible for all statistical data they generate.</u> The confirmation of the absence of any erroneous calculations in new software programs used in the laboratory should be a standard procedure in all laboratories. This process should also include the evaluation of programs that come with automated clinical analyzers, which provide statistical calculation capabilities, and the evaluation of programs used in laboratory information systems.

Special care is recommended when performing statistical calculations with spreadsheet programs. Errors can occur in the following situations:

- When a number is accidentally entered in the area where calculations will occur and then is removed without erasing the entire block, the program may count this erroneous value in computing "N" even when the space appears blank. This is a (@COUNT) problem to be aware of with spreadsheet programs.

- With the significant figures of one's data: While one's spreadsheet calculations may show only 2 places of significant decimal positions, the actual computation that has occurred may be carried to 12 to 14 positions. Use the spreadsheet function labeled @ROUND (cell number, number of places) to obtain a consistent number of decimal places.

- Check to determine if the wrong statistical function is being calculated (population σ, instead of sample s; N−1 in the divisor). Newer spreadsheet versions from commercial software manufacturers have added the terms @STDS and @VARS to generate standard deviation and variance from sample data. If one uses an older program that generates the population standard deviation, one can use the formula listed in Appendix B to convert the population standard deviation value to the sample standard deviation.

Several software manufacturers have recognized the significance of these problems and have integrated solutions into their software programs.

5.0 SUMMARY

This document provides the following for the user of statistics:

- Background information and precautionary notes concerning software used to perform statistical evaluations in the clinical laboratory.
- Test data sets (see Appendix A) that can be used by laboratories to validate the capabilities of software programs in generating statistical results. Type the data from each set into a computer program and determine if the results obtained are the same as those listed. If the results are not the same, then the statistical program being used is generating the wrong result for standard deviation.

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***NOTE:** One special reference review article stands out for tracing out specific statistical tests and recommendations of national committees. This article lists 147 references and was updated in 1993, so the cited reference is current. Every laboratorian should at least have a copy of this 5-page article with all of its references for looking up specific statistical needs.

APPENDIX A

Test Data Sets for Validating Software Calculations of Sample Statistics

Following are two data sets with correctly computed mean, standard deviation, and coefficient of variation for use in validating software calculations of sample statistics. The statistic for standard deviation is calculated using (N−1) degrees of freedom in the denominator.

Data Set #1

Enter these values for "x" in your program:

1
2
3
4
5

and your results will be:

	Mathematical	Computer Rounded
Mean	3.00	3.0
Standard deviation	1.58	1.6*
Coefficient of variation	52.70%	52.7%

Data Set #2

Enter these values for "x" in your program:

3.52
3.26
3.39
3.78
3.93

and your results will be:

	Mathematical	Computer Rounded
Mean	3.5760	3.576
Standard deviation	0.2759	0.276
Coefficient of variation	7.715%	7.72%

*Standard deviation should have no less than two significant figures.

APPENDIX B

Data Conversion Formula

To convert s data calculated using N to data based on N–1, use the following formulation:

$$s(N-1) = s(N) * \mathrm{sqrt}\,[\,N/(N-1)]$$

RELATED NCCLS PUBLICATIONS

EP5-T2 PRECISION PERFORMANCE OF CLINICAL CHEMISTRY DEVICES–SECOND EDITION; TENTATIVE GUIDELINE(1992). EP5-T2 offers guidelines for designing an experiment to evaluate the precision performance of clinical chemistry devices; recommendations on comparing the resulting precision performance claims and determining when such comparisons are valid; and manufacturer's guidelines for establishing claims.

EP6-P EVALUATION OF THE LINEARITY OF QUANTITATIVE ANALYTICAL METHODS; PROPOSED GUIDELINE(1986). EP6-P contains a method for evaluating whether an instrument or quantitative analytical method meets the manufacturer's linearity claim. It also offers guidelines for manufacturer's use when stating a claim of an assay's linear range.

EP7-P INTERFERENCE TESTING IN CLINICAL CHEMISTRY; PROPOSED GUIDELINE(1986). EP7-P contains background information and procedures for characterizing the effects of interfering substances on test results.

EP9-T METHOD COMPARISON AND BIAS ESTIMATION USING PATIENT SAMPLES; TENTATIVE GUIDELINE(1993). EP9-T discusses procedures for determining relative bias between two methods or devices and design of a method-comparison experiment using split patient samples, and analysis of data.

EP10-T2 PRELIMINARY EVALUATION OF QUANTITATIVE CLINICAL LABORATORY METHODS–SECOND EDITION; TENTATIVE GUIDELINE(1993). EP10-T2 discusses experimental design and data analysis for preliminary evaluation of the performance of an analytical method or device.