R13

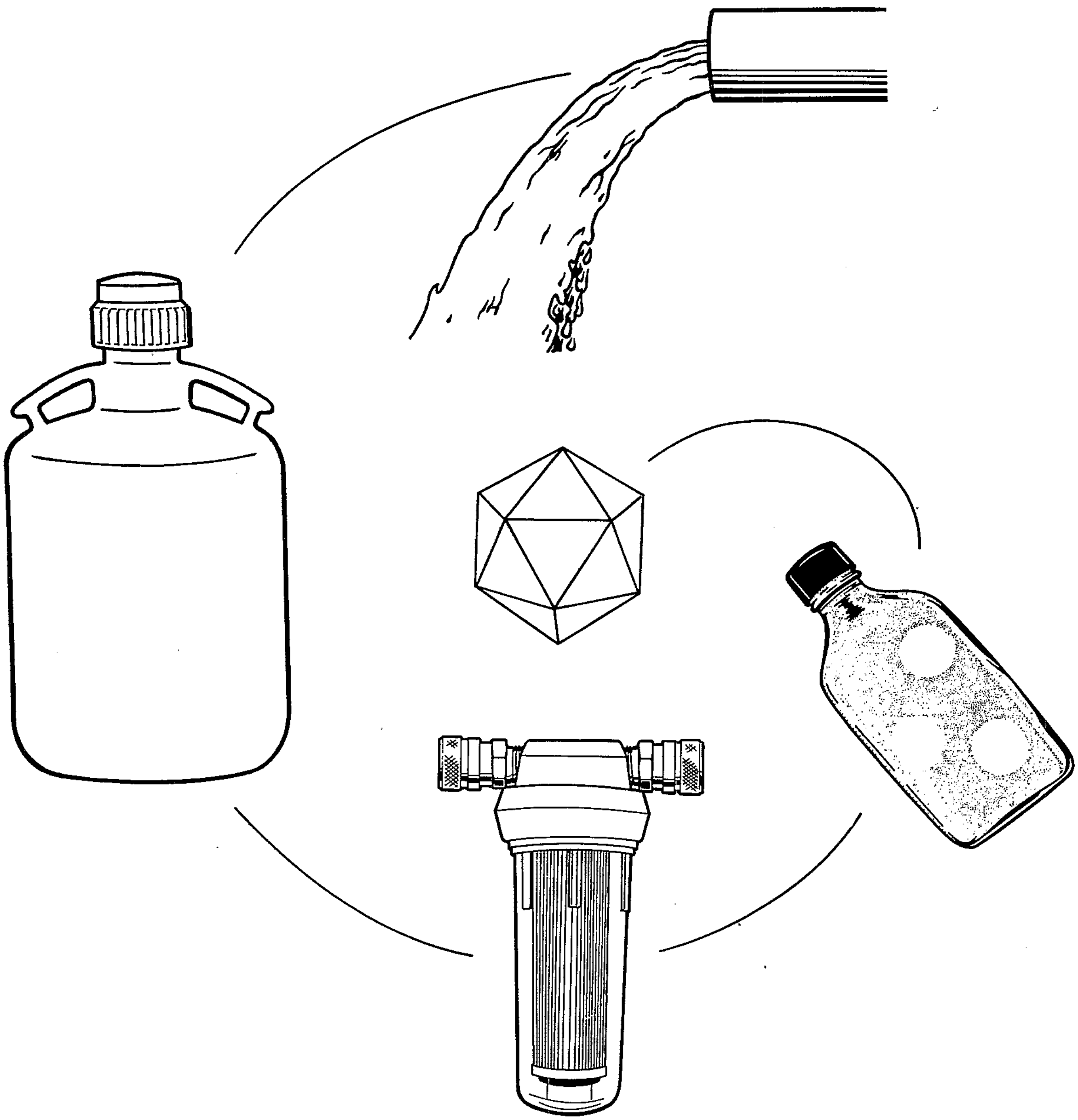
United States
Environmental Protection
Agency

Office of Research and
Development
Washington, DC 20460

EPA/600/4-84-013 ~~(N13)~~
September 1993
Revision

USEPA Manual of Methods for Virology

Chapter 13
September 1993



Chapter 13
Data Analysis and Experimental Design

Prepared by
Larry J. Wymer and Manohari Sivaganesan
Computer Sciences Corporation, Cincinnati, Ohio

1. Introduction

1.1 This chapter provides descriptions and examples of methods for the statistical evaluation of data from virus plaque counting assays. The primary emphasis is on techniques for evaluating the precision of estimates obtained from such assays, in particular, titer estimates of total plaque forming units (PFU), PFU titer by type of virus, and relative frequencies of virus types. Also presented are techniques which should prove useful in the area of quality assurance, namely, methods for evaluating dispersion of the plaques, examining differences between subsets of plaque counts, screening the data for outliers and a discussion of sample design, based on the concepts of precision presented in the previous sections.

1.2 The final section of this chapter includes an extensive compilation of statistical tables designed to facilitate these analyses. With the exception of Table 13-9, these tables have been newly compiled using Statistical Analysis System (SAS) version 5.18 on an IBM 3090 computer. The source for Table 13-9 is ASTM Standard Practice E-178 on dealing with outlying observations (1980).

1.3 The following notations are used uniformly throughout this chapter:

- X the total number of plaques enumerated in all cell culture bottles
- n the total number of cell culture bottles used
- x_i the number of plaques enumerated in the i^{th} cell culture bottle, where the value of the index, i, ranges from 1 through n
- V the total volume of eluate inoculated in all cell culture bottles combined
- v_i the volume of eluate inoculated in the i^{th} cell culture bottle, $i = 1,2 \cdots n$
- t the estimated titer (PFUs per unit volume) of the eluate, $t = X/V$
- k the number of different types of viruses identified in the assay
- X_h the total number of plaques enumerated in all cell culture bottles, identified as being virus type h, $h = 1,2 \cdots k$
- x_{ih} the number of plaques enumerated in the i^{th} cell culture bottle, $i = 1,2 \cdots n$, and identified as being virus type h, $h = 1,2 \cdots k$
- t_h the estimated titer (PFUs per unit volume) of the eluate for the h^{th} virus type,

$$t_h = X_h/V, \qquad h = 1,2 \cdots k$$

The following relationships apply:

$$X = \sum_{i=1}^{n} X_i = X_1 + X_2 + \cdots + X_n$$

$$X = \sum_{h=1}^{k} X_h = X_1 + X_2 + \cdots + X_k$$

$$X_i = \sum_{h=1}^{k} x_{ih}$$

$$V = \sum_{i=1}^{n} V_i = V_1 + V_2 + \cdots + V_n$$

Volumes are expressed in terms of eluate volume; however, these may be converted into equivalent sample volumes without affecting the validity of the analyses.

For example, suppose 10 L of sample is concentrated into 40 mL of eluate of which 1 mL is inoculated in each cell culture bottle. In this case, v_i represents 1 mL, the same for all values of the index, i; if 20 cell culture bottles are inoculated in all (n=20), then V represents 20 mL of eluate. Also, 1 mL of eluate represents 0.25 liter equivalents (Leq) of sample (10 L / 40 mL = 0.25 Leq per mL); therefore, for this example, results of analyses of titer in terms of PFUs per mL of eluate may be converted to PFUs per Leq of sample volume by dividing by a factor of 0.25.

2. Test for Random Dispersion of Plaques

2.1 Introduction

2.1.1 Many of the statistical methods presented in this chapter are based on the assumption that plaques enumerated in the assay are randomly dispersed among cell culture bottles.

2.1.2 This section presents methods for testing the validity of this assumption. In cases where the assumption of randomness is shown to be invalid, alternative methods for analysis of the data are available and included in the appropriate section. Precision of the assay will be greater when the plaques are randomly dispersed, rather than occurring in clusters. In addition, a lack of randomness may indicate a potential problem with the assay, such as contamination of cell lines, or other non-random sources of error.

2.2 Formulas and test statistic

2.2.1 The index of dispersion (Fisher, 1950) is used to test for a random dispersion and is given by

$$D = \sum_{i=1}^{n} \frac{(x_i - t \cdot v_i)^2}{t \cdot v_i}$$

where

- n = total number of cell culture bottles used
- x_i = number of plaques in the i^{th} cell culture bottle;
- i = $1,2,3 \cdots n$
- v_i = eluate volume used in the i^{th} cell culture bottle
- t = estimated PFU titer =

$$\frac{\sum_{i=1}^{n} x_i}{\sum_{i=1}^{n} v_i} = \frac{X}{V}$$

2.2.2 When the same eluate volume is used for each cell culture bottle ($v_i = v$ for $i = 1,2,3 \cdots n$) calculation of the index of dispersion simplifies to

$$D = \sum_{i=1}^{n} (x_i - \bar{x})^2 \,/\, \bar{x} = (n-1)\; S^2 \,/\, \bar{x}$$

where $\bar{x}$ is the average plaque count per bottle,

$$\bar{x} = \sum_{i=1}^{n} x_i/n$$

and s^2 is the sample variance, given by

$$S^2 = \sum_{i=1}^{n} (x_i - \bar{x})^2 / (n-1)$$

$$= \frac{\sum_{i=1}^{n} x_i^2 - \frac{1}{n} x^2}{n-1}$$

the latter expression for s^2 being more convenient for hand calculation.

2.2.3 The test statistic, D, is compared to the 0.05 critical value for chi-square with n-1 degrees of freedom ($\chi^2_{n-1,.05}$). The null hypothesis that the plaques are randomly distributed is accepted when $D < \chi^2_{n-1,.05}$. Table 13-3 gives 0.05 critical values for χ^2 for degrees of freedom ranging from 1 through 50.

In order for this test to be valid, none of the expected plaque counts ($t \cdot v_i$) should be less than one. In addition, no more than 20% of the expected counts should be less than five (Cochran, 1954). When these conditions are not satisfied in the raw data, plaque counts from consecutively numbered cell culture bottles, starting with the highest dilutions, must be combined until these conditions are satisfied by the resulting aggregated data. When data are grouped in this manner, the counts from the combined data are re-indexed; x_i and v_i now refer to the combined counts and volumes, respectively, and n becomes the number of groups, counting any combined data as a single group. If necessary, for example when the expected plaque count from the combined cell culture bottles at the highest dilution level is still less than one, data may be combined among cell culture bottles which have been inoculated with different volumes of eluate.

2.3 Sample calculations

2.3.1 Dataset I is summarized as follows:

bottle number (i)	*eluate per volume bottle (v_i)*	*plaque counts (x_i)*
1-10	*0.001 mL*	*(all zero)*
11-20	*0.01 mL*	*0,2,2,0,2,0,0,2,0,0*
21-30	*0.1 mL*	*10,12,10,6,16,13,9, 14,6,17*

2.3.2 Steps for performing test for random dispersion (Dataset I):

(a) Calculate the expected plaque count for each cell culture bottle.

$E(x_i) = t \cdot v_i$, where

t = total plaque count / total volume of inoculum used

$= \frac{X}{V}$

For this experiment, X = 121 and V = 1.11 mL, giving t = 121/1.11 = 109 PFU/mL. Expected counts for each bottle are, therefore,

bottle numbers	*v_i (mL)*	*Expected count per bottle ($t \cdot v_i$)*
1-10	*0.001*	*109 x .001= 0.109*
11-20	*0.01*	*109 x .01 = 1.09*
21-30	*0.1*	*109 x .1 = 10.9*

(b) If necessary, group the data so that the expected count for each group is at least one and no more than 20% of the groups have an expected count of less than five. In this example, the ten bottles at the highest dilution (v_1-v_{10} = 0.001 mL) have expected counts (0.109) of less than one, and the expected count for each of the ten bottles at the next highest dilution (v_{11}-v_{20} = 0.01 mL) is less than five (1.090). Combining the data from cell culture bottles numbered 1 through 10, 11 through 15, and 16 through 20 results in 13 groups with expected and observed counts as given in columns 4 and 5, respectively, in Table 13-1.

(c) Calculate the individual terms of the index of dispersion (D) from the observed and expected count for each cell culture bottle, or group of cell culture bottles if any data are combined. Individual terms of D are calculated from $(x_i - t \cdot v_i)^2/(t \cdot v_i)$, where i ranges from 1 through 13 for this example. Results of these calculations are shown in column 6 of Table 13-1.

(d) Compare the D statistic, from the summation of the individual terms in step 3 to the appropriate critical value for chi-square. For this example, n = 13; therefore, the appropriate 0.05 critical value for χ^2 with n-1 = 12 degrees of freedom is 21.026, from Table 13-3. Since the computed value of D = 15.413 is less than 21.026, we accept the data as being randomly distributed.

2.3.3 Sample calculations for Dataset II are shown in Table 13-2. Note that only a single dilution of the eluate was used in this experiment, and that no grouping of the data was necessary, since expected plaque counts were greater than five for each cell culture bottle. Otherwise, steps in the calculation of D are identical to those in Section 2.3.2. Because equal volumes of eluate were inoculated in each cell culture bottle, the simplified formula for D given in Section 2.2.2 may be used. Because the value calculated for D (32.530) is greater than the critical value of χ^2 with 13 degrees of freedom (22.362, from Table 13-3), the hypothesis that the plaques are randomly dispersed is rejected.

2.4 Rationale for the test

2.4.1 A "random dispersion of PFUs" means that the probability of finding a PFU in some small volume of the liquid depends only on the volume itself and is not affected by any known presence or absence of PFUs in neighboring regions of the medium. In other words, knowledge that a given region of the eluate contains one or more PFUs would not be useful in identifying other regions where PFUs are most likely to be found. This is a formal statement of what is meant by saying that the eluate is "well mixed."

2.4.2 When PFUs are randomly dispersed, the probability of finding any particular number of PFUs in a fixed volume of the liquid is given by the *Poisson probability distribution* *function* (pdf). The variance of counts that follow a Poisson pdf is equal to their mean. The index of dispersion, D, is simply the ratio of the sample variance to the sample mean of the counts, times n-1; therefore, when the counts follow a Poisson pdf, one would expect the value of D to be near n-1. Because the sample mean and variance used in calculating the index of dispersion will vary from their true values, rarely will the value of D be exactly equal to n-1 even when the data are randomly dispersed.

2.4.3 In those cases in which the counts are not Poisson distributed, the departure from random dispersion is likely to be a result of factors which increase the variance of the counts; thus, when counts are non-Poisson, we expect the test statistic, D, to be greater than n-1. The upper 0.05 critical value for chi-square with n-1 degrees of freedom (Table 13-3) establishes an upper limit for D, beyond which it is not likely (probability < 5%) that such a large value for D could have resulted from counts which follow a Poisson pdf. The test is said to have a *critical level* (sometimes referred to as the alpha level) of 0.05.

2.4.4 When it becomes necessary to combine data because some of the expected plaque counts are less than one or because more than 20% of the expected plaque counts are less than five, any arbitrary grouping of the data is permissible. However, in no case should data be combined on the basis of *observed* plaque counts; for instance, intentionally grouping data from cell culture bottles which contain few plaques specifically with data from cell culture bottles containing a greater number of plaques may invalidate the test for randomness. The recommendation to combine results from consecutively numbered cell culture bottles is made in order to remove, or at least minimize, any influence that the observed plaque counts may have one's decision with regard to any necessary grouping of the data.

2.5 Implications for virus assays

2.5.1 When the index of dispersion (D) test leads to the conclusion that the plaque counts are not randomly distributed, there are two possibilities to consider; the PFUs are not randomly dispersed throughout the medium, or the process of assaying for PFUs has introduced extraneous variability to the results.

In the first case, because the PFUs themselves are not randomly dispersed, one would not expect the resulting plaques to be randomly dispersed. The second possibility may imply that assay conditions were "out of control."

2.5.2 Figure 13-1 illustrates two extreme examples of non-random plaque dispersions (a and b) along with a simulation of a completely random dispersion (c). Each of the three large squares represents the total volume of inoculum; the smaller squares represent those portions of the total volume which are inoculated into individual cell culture bottles. In Figure 13-1(a) the spacing among PFUs is seen to be too uniform to be considered random; in fact, this figure was generated from a uniform lattice of foci with small, random perturbations about each lattice point. On the other hand, plaques are seen to form three distinct clusters in Figure 13-1(b), which was generated from a probability distribution of points about three central locations. As a result, plaques tend to be evenly distributed among all nine cell culture bottles in case a (uniform), while most of the plaques appear in only four of the bottles in case b (clustered). Indices of dispersion for the three cases are calculated to be 1.00 (a: uniform case), 50.75 (b: clustered), and 6.75 (c: random), with eight degrees of freedom each (n = 9). The 0.05 critical value for χ^2 with eight degrees of freedom is 15.507, from Table 13-3.

2.5.3 A uniform distribution of PFUs, such as simulated in Figure 13-1(a), would occur if PFUs exhibited a tendency to repel one another. While such a tendency may not be unreasonable, the notion that PFUs would interact over a range sufficient to affect their behavior on a large scale is not so reasonable. In addition, and most importantly, a uniform distribution is so seldom observed that one feels reasonably assured in discounting any experiment in which it occurs as simply representing an unusual result from a random mixture. Clustering, as simulated in Figure 13-1(b), however, may occur when PFUs are mutually attracted to (or repelled from) specific locations in the liquid. This is not at all unreasonable; areas of net positive or negative ion charge, for instance, might have the large scale effects necessary to produce clustering. Most observed deviations from a random distribution will be consistent with clustering effects.

2.5.4 Procedures are presented in the following sections for determining precision of the estimated titer for both Poisson and non-Poisson distributed plaque counts. If the data are non-Poisson because of variability introduced by the assay procedure, however, the resulting estimate is likely to be biased. Unfortunately, it is not usually possible to distinguish between non-random dispersion of PFUs and systematic biases resulting from the assay procedure solely by means of statistical analysis of the data. Statistical procedures are presented in Sections 3 and 4, however, which may be useful for evaluating non-random counts under certain circumstances. Occasionally, it may not be possible to perform the test for a random dispersion on a given set of experimental data. This would be the case whenever fewer than ten plaques are observed in total from among all cell culture bottles used in the assay. In such cases, while the assumption of a random dispersion cannot be verified, the recommendation, nevertheless, is to proceed with this assumption in the ensuing statistical evaluation.

3. Tests for Outliers

3.1 Introduction

3.1.1 An outlier is an observation that is discordant in relation to the other sample data. In the context of virus plaque count assays, an individual plaque count which appears to be unusually high or low compared to the other counts may be considered an outlier.

3.1.2 Statistical procedures are presented in this section for evaluating the hypothesis that an apparent outlying observation is merely a result of the inherent variability of the data, rather than representing a truly aberrant value as a result of some discrepancy in the way in which that observation was obtained. When these procedures indicate that an observation is a true outlier, this should not be construed to imply that such an observation be automatically discarded. While any datum found to be, statistically, an outlier should be noted in reporting the results of the assay, the decision to exclude the observation must be based on, at a minimum, the researcher's opinion that the observation is likely to have been in error.

3.2 Statistical tests for outliers

3.2.1 The test to be used in evaluating whether an observation (or two observations) may be considered an outlier depends on whether the remaining observations, excluding the potential outlier(s), are randomly distributed. The test for a random distribution of plaque counts, as presented in Section 2 may be performed on plaque counts excluding those cell culture bottles yielding the suspected outlier counts. However, if the test of randomness performed on all the plaque counts, including the suspected outlier(s), indicates that the assumption of a random dispersion is reasonable, it is not necessary to retest the subset of counts excluding the outlier(s).

Because the tests for outliers from a random distribution are simple to perform, involving no computation other than totalling all plaque counts, the recommendation is to perform a test on the assumption of randomness for the remaining counts only if the test for an outlier from a random distribution indicates that the value in question is significant. If the observation is not an outlier based on the assumption of randomness, neither will it be a statistical outlier with this assumption removed.

3.2.2 Tests are presented for four potential outlier situations:

(a) a single apparent high value
(b) a single apparent low value
(c) a pair of apparent upper outliers
(d) a pair of apparent lower outliers

3.2.3 Tests for outliers: randomly dispersed data

(a) For a single upper or lower outlier, the test statistic consists of the value itself, conditional on the total plaque count in all cell culture bottles at the same dilution of eluate. For a pair of upper or lower outliers, the test statistic becomes the sum of the two extreme counts, conditional on the total count. Descriptions of these tests may be found on pages 198-200 in Barnett and Lewis (1984).

(b) To test for a single *upper* outlier, determine the greatest number of plaques obtained in a single bottle from among all cell culture bottles inoculated with identical volumes of eluate. Enter Table 13-4 at the row corresponding to this number and the column corresponding to the number of cell culture bottles used. If the total of all plaques enumerated in all cell culture bottles (including the value in question) is *lower* than the entry in Table 13-4, then the observation in question may be considered to be an outlier at the 0.01 critical level.

(c) Table 13-5 indicates *lower* outlier values. Enter this table at the row corresponding to the lowest count among all cell culture bottles inoculated with the same volume of eluate. If the total plaques enumerated in all cell culture bottles (including the value in question) is *greater* than the entry under the column for the appropriate number of cell culture bottles, then the observation in question may be considered a lower outlier at the 0.01 critical level.

(d) Critical values for upper and lower outlier *pairs* are indicated in Tables 13-6 and 13-7, respectively.
These tables are used in a manner similar to their single outlier counterparts (Tables 13-4 and 13-5), except that the rows correspond to the *sum* of the two highest or two lowest counts, respectively.

3.2.4 Tests for outliers: non-randomly dispersed data

(a) Order the counts from n cell culture bottles from lowest to highest, so that

$$x_1 \le x_2 \le x_3 \le \cdots \le x_{n-1} \le x_n$$

Compute the variance of the counts:

$$s^2 = \sum_{i=1}^{n}(x_i - \bar{x})^2 / (n-1)$$

$$= \frac{\sum_{i=1}^{n} x_i^2 - \frac{1}{n}X^2}{n-1}$$

where $\bar{x}$ is the sample mean of the counts,

$$\bar{x} = \frac{1}{n}\sum_{i=1}^{n} x_i.$$

(b) For a single *upper* outlier, compute the statistic,

$$T_n = \frac{(x_n - \bar{x})}{s}$$

where s, the sample standard deviation, is the square root of s^2, the sample variance as given above, in Section 3.2.4 Step (a).
Compare the resulting value of T_n with the appropriate entry in Table 13-8 for the total number, n, of cell culture bottles used. If T_n is greater than or equal to this entry, then the count, x_n, may be considered an upper outlying value. The procedure for testing a *lower* outlier is similar, except that the appropriate test statistic is given by,

$$T_1 = \frac{(\bar{x} - x_1)}{s}$$

Table 13-8 gives the minimum values for T_1 such that x_1 may be considered a lower outlying value.

(c) Given a *pair* of potential lower outliers, the following test statistic may be used,

$$T_{1,2} = SS_{1,2} / SS$$

where SS is the total sum of squares, $SS = (n-1)s^2$, and $SS_{1,2}$ is the sum of squares omitting the two lowest observations,

$$SS_{1.2} = \sum_{i=3}^{n}(x_1 - \bar{x}_{1,2})^2$$

The sample mean of the remaining counts, $\bar{x}_{1,2}$, is given by

$$\bar{x}_{1,2} = \sum_{i=3}^{n} x_i / (n-2)$$

Table 13-9 gives the critical values for T such that $T_{1,2} < T$ indicates a statistically significant lower outlier pair. For a pair of apparent upper outlier values, use the statistic,

$$T_{n-1,n} = SS_{n-1,n} / SS$$

where

$$SS_{n-1,n} = \sum_{i=1}^{n-2}(x_1 - \bar{x}_{n-1,n})^2.$$

and $\bar{x}_{n-1,n}$ is the sample mean of the remaining counts,

$$\bar{x}_{n-1,n} = \sum_{i=1}^{n-2} x_1 \;/\; (n-2)$$

Again, Table 13-9 gives the maximum values for $T_{n-1,n}$ for the pair to be considered as representing two upper outlying values.

3.3 Example

3.3.1 Data for this example consist of the following set of plaque counts from ten cell culture bottles, all inoculated with 0.1 mL of sample eluate:

8, 12, 7, 4, 6, 11, 10, 5, 5, 19

3.3.2 The procedure for performing the test for outliers is

(a) Identify apparent outlying observations.

In this case, the last count in the series, 19, appears to be high relative to the other values.

(b) Find the total of all plaques in the series.

For these data, the total number of plaques enumerated is 87.

(c) Compare the total from Step (b) with the entry from the appropriate table (Table 13-4, 13-5, 13-6 or 13-7), depending on the direction and number of suspected outliers.

One potentially high value was identified; therefore, Table 13-4 is appropriate for this example. The total count (87) from step 2 is less than the tabulated value (89) for a high count of 19 among 10 cell culture bottles; therefore, this observation is considered to be a statistical outlier. This conclusion is, however, conditional on whether the remaining data are randomly dispersed.

(d) If steps 1-3 above indicate that the observation(s) is an outlier, recompute the D statistic (Section 2) to perform the test for randomness on the remaining observations.

Omitting the count of 19 from these data, and following the steps outlined in Section 2.3.2, Step (d) is recomputed to be 8.765. This is less than the 0.01 critical value for Chi-square with 8 degrees of freedom (= 15.507 from Table 13-3); therefore, we accept that the remaining data are randomly distributed. This validates the finding that the result of 19 plaques in one cell culture bottle represents a statistically significant outlier.

3.4 Rationale for the test

3.4.1 Tests for outliers using Tables 13-4 through 13-7 consider the results to represent a multinomial distribution of plaques among cell culture bottles, where a plaque may appear in any of the cell culture bottles with equal probability. The multinomial distribution function gives the probability for any given arrangement of n objects (plaques) among m containers, each object being *randomly* and independently assigned to one container. Given this distribution, the probability that the number of objects assigned to a single container (or two containers in the case of a pair of outliers) will exceed, or fall below a given value can be calculated. The values in Tables 13-4 through 13-7 are such that this probability is 1% or less. The test for outliers, therefore, is said to have a *critical level* of 1%; use of the 1% critical level is in accordance with the American Society for Testing and Materials (ASTM) standard practice E-178 (1980) for dealing with outlying observations.

For further discussion of outliers from Poisson samples, see Barnett and Lewis (1984).

3.4.2 Tests for outliers based on Tables 13-4 through 13-7 assume that the plaques are randomly (Poisson) distributed among cell culture bottles, with the possible exception of the outlying observation(s) itself. The index of dispersion is recomputed for the remaining data to test this assumption. When the remaining data fail the test for randomness, these outlier tests are not valid.

3.4.3 Alternative tests for outliers when the random dispersion assumption is shown to be invalid, given in Section 3.2.4 and using Tables 13-8 and 13-9, assume that plaque counts are normally distributed with unknown mean and variance, and are based on ASTM standard practice E-178 (1980) for dealing with outlying observations.

Unless the data are approximately normally distributed, the actual critical levels associated with these tests will differ from the nominal level of 1%. The requirements for statistical outliers based on Tables 13-8 and 13-9, however, are more stringent than the requirements for statistical outliers from a random distribution. The outliers have to be more extreme, because of the assumption that the variance of non-randomly dispersed plaque counts will be greater than that of randomly dispersed plaque counts.

3.5 Implications for virus assays

3.5.1 Some potential reasons for outlying observations among viral plaque count data include

(a) Contamination of cell culture bottles

(b) Errors in inoculation

(c) Variation in incubation conditions

(d) Counting errors.

3.5.2 The above conditions, among others, will produce outliers only when they differentially affect only one or two observations. If, for example, all cell culture bottles become contaminated, this may be evidenced by an increased variance among plaque counts (non-random counts) or by a non-detectable bias, but the observations may still be concordant among themselves.

3.5.3 When an aberrant condition is known to have affected the results in one or more of the cell culture bottles, these data should be discarded without regard to whether they appear to be outliers. Tests for outliers are inappropriate when there are known reasons for invalidating data.

3.5.4 When an observation is shown to be an outlier, an effort should be made to discover any variance in the conditions under which that observation was obtained. In practice, however, such efforts often prove to be fruitless; thus, the decision whether to reject the observation in question becomes a matter for the researcher's judgement. At a minimum, the existence of statistical outliers should be noted in reporting the assay results. When outlying observations are discarded, the researcher's reasons for doing so, beyond their being statistically significant outliers, should be stated.

4. Test for Differences Between Groups of Plaque Counts

4.1 Introduction

4.1.1 This section presents methods for comparing results between two sets of plaque counts.

4.1.2 Groupings may be defined in any logical manner, such as two different dilutions in a series, different cell culture lines, or different methods of incubation.

One would never expect the results between two such groupings to agree perfectly, even when there is known to be no real difference between the groups themselves, simply because of random sampling variation. The purpose of statistical analysis of the difference is to determine whether one might reasonably attribute the difference to chance.

4.2 Test statistic

4.2.1 Calculate the PFU titer estimate for each group:

$$t_i = \frac{X_i}{V_i}$$

where X_i is the total plaque count in all cell culture bottles in group i (i=1,2) and V_i is the total volume of eluate with which group i was inoculated. Designate the groups as group 1 and group 2 so that $t_1 < t_2$. Calculate the ratio (Cox, 1953),

$$R = \frac{t_2 + \frac{1}{2V_2}}{t_1 + \frac{1}{2V_1}} = \frac{\left(X_2 + \frac{1}{2}\right) / V_2}{\left(X_1 + \frac{1}{2}\right) / V_1}$$

4.2.2 When equal volumes of eluate are used in each group ($V_1 = V_2$), the ratio simplifies to

$$R = \frac{X_2 + \frac{1}{2}}{X_1 + \frac{1}{2}}$$

In this case, the index 1 refers to the group with the lower total plaque count, X_1.

4.2.3 The test statistic, R, is compared to the 0.05 two-tailed critical value for the F distribution with $2 \cdot X_1 + 1$ degrees of freedom for the numerator, and $2 \cdot X_2 + 1$ degrees of freedom for the denominator. Table 13-10 gives 0.05 two-tailed critical values for the F distribution.

4.2.4 When equal volumes are used in each group ($V_1 = V_2$), Table 13-11 can be used to determine the minimum value for the higher plaque count (X_2), based on the observed value of the lower count (X_1). Note that in this case the critical value for F is uniquely determined by the corresponding value for X_2, given X_1.

4.2.5 A 95% confidence interval for the ratio of PFU titers (higher PFU titer/ lower PFU titer) may be constructed by

$$(R \cdot F-,\ R \cdot F+)$$

where F+ and F- are the upper and lower 2.5% critical values of F with ($2X_2+1$, $2X_1+1$) degrees of freedom.

Note that when the F test does not show a significant difference between the two groups, this interval will include the value 1. Moreover, a 95% confidence interval shows the degree of uncertainty in the comparison and it makes it easier to asses the practical significance of a difference.

4.3 Examples

Unequal volumes of eluate are used in example 1 and equal volumes of eluate are used in example 2.

4.3.1 Example 1

Data for this example consist of plaque counts from ten cell culture bottles at each of two dilutions of eluate as shown below. A test for difference between the two dilutions is to be performed.

bottle numbers	eluate volume per bottle (v_i)	plaque counts
1 thru 10	0.01 mL	5,4,4,6,8,3,5,9,3,7
11 thru 20	0.1 mL	31,37,42,31,38, 27,30,27,26,45

4.3.2 Steps for performing the test for difference are

(a) Calculate the titer estimate for each group separately. This is the total plaque count for the respective group divided by the total volume of eluate used in that group. For the example, among the 0.01 mL bottles, $t_{(.01\,mL)} = 54/\,0.1 = 540$ PFU/mL, and for the 0.1 mL bottles, $t_{(.1\,mL)} = 334/1.0 = 334$ PFU/mL.

(b) Identify the group with the lower PFU titer, label it group 1, and compute the ratio, R as given in Section 4.2.1. For this example,

$$R = \frac{540 + \frac{1}{2 \cdot 0.1}}{334 + \frac{1}{2 \cdot 1.0}} = \frac{\left(540 + \frac{1}{2}\right) / 0.1}{\left(334 + \frac{1}{2}\right) / 1.0}$$

$$= 1.629$$

(c) Compare the value of R from step 2 with the 0.05 critical value of the F distribution with $2 \cdot X_1 + 1$ and $2 \cdot X_2 + 1$ degrees of freedom (Table 13-10).

In this case, the numerator and denominator degrees of freedom are, respectively, 669 (= 2 • 334 + 1), and 109 (= 2 • 54 + 1). From Table 13-10, the 0.05 critical value for the F distribution is less than 1.378 whenever the degrees of freedom for the numerator are greater than 500 and the degrees of freedom for the denominator are greater than 100. Therefore, the calculated value for R from step 2, 1.629, clearly exceeds the critical F value, and we may infer that the results between the two dilution levels differ significantly.

4.3.3 Example 2

Data for this example consist of plaque counts from ten cell culture bottles of equal volumes of eluate.

bottle numbers	eluate volume per bottle (v_i)	plaque counts
1 thru 10	0.1 mL	5,4,4,6,8,3,5,4,4,7
11 thru 20	0.1 mL	8,7,4,6,9,5,7,4,5,7

A test for difference between the two groups is to be performed.

4.3.4 Steps for performing the test for difference and for constructing a 95% confidence interval are

(a) Identify the group with the lower PFU titer and label it as group 1.

For this example, the observed lower count X_1 is equal to 50 and the observed higher count X_2 is equal to 62.

(b) Compare the value of X_2 from step 1 with the 0.05 two-tailed critical value M for the larger of the two Poisson counts (Table 13-11).

In this case, the 0.05 critical value M = 72 (for lower count of 50) is greater than the observed higher count 62. This means that there is no significant difference (statistically) between the two groups.

(c) Construct a 95% confidence interval for the ratio (higher PFU titer/lower PFU titer). For example 2

$$R = (2X_2 + 1) / (2X_1 + 1) = (2 \cdot 62 + 1) / (2 \cdot 50 + 1) = 125 / 101 = 1.24$$

The upper 2.5% critical value F+ of F with (2 • X_2 +1 , 2• X_1 + 1) = (125,101) degrees of freedom is 1.459 (Table 3.10). The lower 2.5% critical value of F with (125,101) degrees of freedom is the reciprocal of the upper 2.5% critical value of F with (101,125) degrees of freedom and is equal to 1/1.448 = 0.691. A 95% confidence interval is given by (R • F- , R • F+) = (0.856 , 1.809).

4.3.5 Occasionally, interpolation of the values in Table 13-10 may be necessary in order to determine whether a calculated value for R is statistically significant. In such cases, *inverse interpolation* on degrees of freedom is recommended; this requires taking the inverses of the tabulated degrees of freedom and desired degrees of freedom before performing the actual interpolation. While interpolation is not necessary in the example given in this section, because the calculated value for R clearly exceeds any possible critical F-value, the appropriate critical F-value may be estimated as follows:

(a) The entries in Table 13-10 which bracket the required numerator and denominator degrees of freedom (669 and 109, respectively) are

Denominator degrees of freedom: (ddf)	Numerator degrees of freedom (ndf): 500	1000
100	1.378	1.363
150	1.307	1.290

(b) Interpolate the critical F-values for numerator degrees of freedom equal to 669. For this interpolation, use the multiplier:

$$m1 = \frac{\frac{1}{669} - \frac{1}{500}}{\frac{1}{1000} - \frac{1}{500}} = 0.505$$

Perform this interpolation for denominator degrees of freedom (ddf) = 100 and 150:

(b.1) For ddf = 100:

$$F_{669,100} = 1.378 + m1 \cdot (1.363 - 1.378) = 1.378 + 0.505 \cdot (-0.015) = 1.378 - 0.008 = 1.370$$

(b.2) For ddf = 150:

$$F_{669,150} = 1.307 + m1 \cdot (1.290 - 1.307) = 1.307 + 0.505 \cdot (-0.017) = 1.307 - 0.009 = 1.298$$

(c) Interpolate the critical F-values for denominator degrees of freedom equal to 109. For this interpolation, use the multiplier:

$$m2 = \frac{\frac{1}{109} - \frac{1}{100}}{\frac{1}{150} - \frac{1}{100}} = 0.248$$

Use the F-values found in Step (b):

$$F_{669,109} = 1.370 + m2 \cdot (1.298 - 1.370) = 1.370 + 0.248 \cdot (-0.072) = 1.370 - 0.018 = 1.352$$

4.4 Rationale for the test

4.4.1 Inverse sampling refers to a sampling technique in which the number of events (plaques) to be observed is fixed in advance; what varies in inverse sampling is the volume required for exactly this number of events to be observed. Under inverse sampling, if X_1 and X_2, as defined in Section 4.2.1, had been specified in advance, and exactly V_1 and V_2 volumes of medium had been required to attain these predetermined plaque counts, then the ratio t_2/t_1 can be shown to follow an F distribution with 2 • X_1 degrees of freedom in the numerator and 2 • X_2 degrees of freedom in the denominator, where $t_j = X_j / V_j$, j=1,2. In our case, because sampling was not stopped at the moment that the X_1^th^ plaque occurred, we conclude that V_1 was most likely to have been greater than would have been necessary for exactly X_1 plaques to form, but less than was necessary for X_1 + 1 plaques to form. Therefore, X_1 + 1/2 is taken as the number of occurrences. The same reasoning applies to X_2, leading to the test statistic,

$$R \doteq \frac{\left(X_2 + \frac{1}{2}\right) / V_2}{\left(X_1 + \frac{1}{2}\right) / V_1}$$

4.4.2 The test for differences between groups of plaque counts, as presented in this section is a two-tailed (or two-sided) test because the group with the higher titer estimate is always placed in the numerator of the test statistic, R. Thus, the test is for any difference between the two groups, regardless of which group may be determined to have the higher titer.

4.5 Implications for virus assays

4.5.1 A statistically significant difference between two groups of plaque counts indicates that the magnitude of the difference is large enough that it is not likely to have occurred by chance. The "critical level" for the test specifies the risk assumed in rejecting the equality between the two groups, when, in fact, the true means for the two groups are equal (a "Type I" or "alpha" error).

A critical level of 0.05 is probably the most commonly used value in most fields of application, although critical levels of 0.01 and 0.10 are also commonly used, depending on the level of risk the experimenter is willing to assume.

4.5.2 The F test may be used whenever one wishes to compare data from two groups. Some examples of comparisons that might be made, in addition to dilution series as in the example of Section 4.3, are susceptibilities of different lines of a cell culture, and inoculations at different points in time.

Comparisons of this sort should be made only between plaques from the same titration, however, since differences detected between plaques obtained from separate titrations may simply reflect differences in the relative recoveries of PFUs between the titrations.

5. Confidence Intervals for PFU Titer

5.1 Introduction

5.1.1 Methods presented in this section deal with the precision of the PFU titer obtained from a single titration viral assay.

5.1.2 The titer calculated from a sample volume of material is a single-valued, or "point", estimate of the PFU titer in the population from which the sample was drawn. An interval estimate, on the other hand, is a range of values such that there is a known probability that the true value of the population parameter, such as titer, will be contained within this range. Commonly, this probability is set at 95%, yielding a 95% confidence interval.

5.1.3 Two point estimates of titer may be identical but have different precisions, as defined by the widths of their respective 95% confidence intervals.

5.2 Procedure

5.2.1 Following the procedure given in Section 2, first determine whether the assumption that the plaques are randomly dispersed is valid. The method used for computing 95% confidence limits will depend on whether the plaques are found to be randomly distributed.

5.2.2 When the data are randomly dispersed, use Table 13-12 for the lower and upper 95% confidence limits for the total number of plaques, based on the total plaques actually enumerated in the sample. Let X represent the total number of plaques in all cell culture bottles, let X_L and X_U represent the lower and upper 95% confidence limits, respectively, from Table 13-12, and let V represent the total volume of eluate (in mL) used in all cell culture bottles. The estimated titer is, then, given by t = X/V, and lower and upper 95% confidence limits for titer are given by X_L/V and X_U/V, respectively.

5.2.3 When the data fail the test for randomness (Section 2), 95% confidence limits for total PFU titer may be approximated by

$$t \pm 1.96\frac{\sqrt{n \cdot s}}{V}$$

where

$$s^2 = \frac{\sum_{i=1}^{n}(x_i - v_i \cdot t)^2}{n-1}$$

is the sample variance of t, and
s, the square root of s^2, is the sample standard deviation of t
n is the number of cell culture bottles used
x_i is the plaque count in the i^{th} bottle, i=1,2 • • • n
v_i is the eluate volume used in the i^{th} bottle
t and V are as defined in Section 5.2.2, above.

When equal volumes of eluate are inoculated (v_i=v, a constant volume), the sample variance (s^2) may be calculated as

$$s^2 = \frac{\sum_{i=1}^{n} x_i^2 - \frac{1}{n}\left(\sum_{i=1}^{n} x_i\right)^2}{n-1}$$

5.3 Examples

5.3.1 For Dataset I (see Section 2.3.1), the assumption that the plaques are randomly dispersed was previously shown to be valid. Titer is estimated as 121 + 1.11 = 109.0 PFU/mL. Table 13-12 gives lower and upper confidence limits of 100.4 and 144.6 for the total plaque count, given the sample count of 121. Therefore, the 95% confidence interval for the true titer is
100.4 + 1.11 = 90.5 PFU/mL to
144.6 + 1.11 = 130.3 PFU/mL.

5.3.2 Plaque counts in Dataset II (Table 2.2) were shown to be non-randomly dispersed. Calculation of the 95% confidence interval for the PFU titer is as follows:

(a) Calculate the titer estimate,

t = X/V = 166/14 = 11.9 PFU/mL

(b) Compute

$$s^2 = \frac{\sum_{i=1}^{n}(x_i - v_i \cdot t)^2}{n-1} = \frac{385.74}{13} = 29.672$$

(c) Compute the quantity

$$\frac{1.96 \cdot \sqrt{n} \cdot s}{V} = \frac{1.96 \cdot \sqrt{14} \cdot \sqrt{29.672}}{14} = \frac{1.96 \cdot 3.742 \cdot 5.447}{14} = 2.9$$

(d) Subtract the result from Step (c) from the result of Step (a) to obtain the lower limit, and add these quantities for the upper limit. The resulting 95% confidence interval is
11.9 - 2.9 = 9.0 PFU/mL to
11.9 + 2.9 = 14.8 PFU/mL.

5.4 Rationale

5.4.1 When the data are randomly distributed, the probability of finding a given number of PFUs in the sample volume of eluate can be calculated by means of the Poisson probability distribution function (pdf). The Poisson pdf depends only on the mean number of PFUs per unit volume. The lower limit for the confidence interval in Table 13-12 is the value of this mean such that the probability of finding at least as many plaques as were actually observed is 2.5%. Likewise, the upper limit is the value of the mean for which the probability of finding as many plaques as were actually observed, or fewer, is 2.5%.

This procedure was originally presented by Garwood (1936).

5.4.2 The alternate procedure for determining a 95% confidence interval for the PFU titer is based on the normal probability distribution approximation to the confidence limits for a ratio estimate (Cochran, 1977: pp 150-165). Regardless of the distribution of the actual data, the sample ratio of PFUs per unit volume, t, will approach a normal distribution as the sample size increases. How large the sample needs to be in order for the normal distribution to approximate the confidence interval reasonably well will vary. In general, a smaller sample will be required for this approximation to be adequate when the number of PFUs per unit volume is relatively large than when there are relatively few PFUs per unit volume.

On pages 39-44, Cochran (1977) discusses the adequacy of the normal approximation for computing confidence limits for the sample mean in general.

5.4.3 Implications for virus assays

(a) The sample volume of material which is assayed is taken to be representative of a larger population of the same material.
Confidence intervals as given in this section pertain to the true value of "recoverable" PFUs per unit volume in the population, assuming assay conditions resulting in identical relative recoveries of PFUs. That is, only single titration, and not multi-titration, precision is considered.

(b) Failure of the titration method to recover all viruses present in the original sample is regarded as a bias.
It is not possible to evaluate this bias for a single titration assay without parallel assay of a known standard.

6. Simultaneous Confidence Intervals for PFU Titer by Virus Type

6.1 Introduction

6.1.1 Simultaneous 95% confidence intervals for titer by each type of virus identified in the sample are calculated in such a way that the probability that all the intervals contain the true values of their respective population parameters is 95%.

6.1.2 For this to be true, confidence intervals for the individual estimates must be wider than they would be for a single confidence interval for total titer, because the probability of error (that a confidence interval will not contain the true value of its respective population parameter) is compounded by the fact that there are more opportunities for error.

6.2 Procedure

6.2.1 As in the procedure for the determination of a single 95% confidence interval for total PFU titer, the method used for computing simultaneous 95% confidence intervals for PFU by type of virus will depend on whether the test for random dispersion of all plaques (see Section 2) has indicated that the assumption of randomness is valid. The method for computing simultaneous confidence intervals, then, will correspond to the method used for determining the confidence interval for overall PFU titer.

6.2.2 When the data are randomly dispersed, use Table 13-13 for the lower and upper 95% confidence limits for the total number of plaques for each type of virus identified in the sample, based on the total plaques of that virus type actually enumerated in the sample and the number of different types identified. Let X_h represent the total number of plaques for the given virus type in all cell culture bottles, let X_{hL} and X_{hU} represent the lower and upper 95% confidence limits, respectively, from Table 13-13, and let V represent the total volume of eluate (in mL) used in all cell culture bottles. The estimated titer is, then, given by $t_h = X_h / V$, and lower and upper 95% confidence limits for titer are given by X_{hL} / V and X_{hU} / V, respectively.

6.2.3 When the data fail the test for randomness (Section 2), 95% simultaneous confidence limits for the PFU titer by virus type may be approximated by

$$t_h \pm \frac{z \cdot \sqrt{n} \cdot s_h}{V}$$

where

$$s_h^2 = \frac{\sum_{i=1}^{n} (x_{ih} - v_i \cdot t_h)^2}{n-1}$$

is the sample variance of t_h, and
s_h (the square root of s) is the sample standard deviation
n is the number of cell culture bottles used
x_{ih} is the plaque count in the i[th] bottle, i=1,2,3 • • • n
v_i is the eluate volume inoculated in the i[th] bottle
t_h and V are as defined in Section 6.2.2, above.

The value for "z" will depend on the number of different types of viruses identified in the sample:

number of virus types	z
2	2.24
3	2.39
4	2.50
5	2.58
6	2.64
7	2.69
8	2.73

When equal volumes of eluate are inoculated (v_i=v, a constant volume), the sample variance (s^2) may be calculated as

$$s_h^2 = \frac{\sum_{i=1}^{n} x_{ih}^2 - \frac{1}{n}\left(\sum_{i=1}^{n} x_{ih}\right)^2}{n-1}$$

$$= \frac{\sum_{i=1}^{n} x_{ih}^2 - \frac{1}{n} X_h^2}{n-1}$$

6.3 Examples

6.3.1 For the plaque counts of Dataset I (Section 2.3.1), the assumption of random dispersion was shown to be valid. The 95% confidence interval for overall titer was calculated in Section 5, and found to be 90 to 130 PFU/mL. Identification of the plaques observed in Dataset I yields the following results:

Type	Number found
Coxsackie virus CB2	67
Coxsackie virus CB5	37
Polio virus PV1	17

Total eluate volume used in all cell culture bottles was 1.11 mL.

6.3.2 Because three virus types were identified in the sample, 95% lower and upper confidence intervals for the number of plaques by type of virus are found from Table 13-13 for "number of groups = 3". These limits are divided by the total eluate volume (1.11 mL) to obtain the 95% simultaneous confidence intervals for titer by type of virus as shown below:

	CB2	CB5	PV1
Number found	67	37	17
Confidence Limits from Table 13-13:			
lower	48.99	24.02	8.717
upper	89.31	54.32	29.71
Overall titer PFU/mL	60	33	15
95% Confidence Interval PFU/mL	44 - 80	22-49	8-27

6.3.3 Plaque counts in Dataset II (Table 13-2) were shown to be non-randomly dispersed. The 95% confidence interval for the overall PFU titer was calculated in Section 5 to be from 9.0 to 14.8 PFU/mL. Identification of the plaques found in Dataset II yields the following results:

Type	Number found in each bottle	Total
CB2	3,6,4,4,9,7,3, 8,4,2,13,5,9,4	81
CB5	2,6,1,3,5,10,6, 6,2,2,4,2,5,2	56
PV1	2,2,2,3,1,4,0, 4,0,2,3,1,3,2	29

6.3.4 Detailed calculations for CB2 virus (h=1) are shown below. Because the same eluate volume is used in each cell culture bottle (1 mL each), the simplified formula for calculating the sample variance, given in Section 6.2.3, may be used.

Bottle (i)	x_{1i}	x^2_{1i}	v_i(mL)
1	3	9	1
2	6	36	1
3	4	16	1
4	4	16	1
5	9	81	1
6	7	49	1
7	3	9	1
8	8	64	1
9	4	16	1
10	2	4	1
11	3	169	1
12	5	25	1
13	9	81	1
14	4	16	1
Total	81	591	14

$$s_1^2 = \frac{591-81^2/14}{14-1} = \frac{591-468.64}{13} = 9.4123$$

$$s_1 = \sqrt{9.4123} = 3.068$$

$$t_1 = 81/14 = 5.8 \text{ PFU/mL}$$

95% lower confidence limit =

$$t_1 - \frac{z \cdot \sqrt{n} \cdot s_1}{V}$$

$$= 5.8 - \frac{2.39 \cdot \sqrt{14} \cdot 3.068}{14}$$

$$= 3.8 \ PFU/mL$$

95% upper confidence limit =

$$t_1 + \frac{z \cdot \sqrt{n} \cdot s_1}{V}$$

$$= 5.8 + \frac{2.39 \cdot \sqrt{14} \cdot 3.068}{14}$$

$$= 7.8 \ PFU/mL$$

6.3.5 Similar calculations are made for the remaining two virus types. 95% simul-taneous confidence intervals for all three types are given below.

Type	Titer	95% Confidence Interval (PFU/mL)
CB2	5.8	3.8 - 7.8
CB5	4.0	2.4 - 5.6
PV1	2.1	1.3 - 2.9

6.4 Rationale

6.4.1 When the data are randomly distributed, the probability of finding a given number of PFUs in the sample volume of eluate can be calculated by means of the Poisson probability distribution function (pdf). The Poisson pdf depends only on the mean number of PFUs per unit volume. The lower limits for the confidence interval in Table 13-13 is the value of this mean such that the probability of finding at least as many plaques as were actually observed is 2.5%/k, where k represents the number of different virus types found in the sample. Likewise, the upper limit is the value of the mean for which the probability of finding as many plaques as were actually observed, or fewer, is 2.5%/k. These are referred to as "Bonferroni adjusted confidence intervals," a discussion of which may be found in Snedecor and Cochran (1980, pp 166-167). The probability that the confidence interval for the titer of any one specific virus type will be incorrect (that is, the interval will not contain the true titer value) is 5%/k; the probability that *one or more* of the k confidence intervals will be incorrect is, according to Bonferroni's inequality, no greater than k • 5%/k = 5%.

6.4.2 The alternate procedure for determining simultaneous 95% confidence intervals for PFU titers is based on the normal probability distribution approximation to the confidence limits for a ratio estimate (Cochran, 1977: pp 150-165). Regardless of the distribution of the actual data, the distribution of each sample ratio (t_h) will approach that of a normal distribution as the sample size increases. How large the sample needs to be in order for the normal distribution to approximate the confidence interval reasonably well will vary. In general, a smaller sample will be required for this approximation to be adequate when the number of PFUs per unit volume for a given virus type is relatively large than when there are relatively few PFUs per unit volume.

6.5 Implications for virus assays

6.5.1 The sample volume of material which is assayed is taken to be representative of a larger population of the same material.

Confidence intervals as given in this section pertain to the true value of "recoverable" PFUs per unit volume in the population, assuming assay conditions resulting in identical relative recoveries of PFUs. That is, only single titration, and not multi-titration, precision is considered.

6.5.2 Failure of the titration method to recover all viruses of any given type present in the original sample is regarded as a bias. It is not possible to evaluate this bias for a single titration assay without parallel assay of a known standard.

6.5.3 Simultaneous confidence intervals are appropriate when information regarding any individual type of viruses identified in the assay is equally important. When the interest centers on one particular virus type, a single 95% confidence interval for the titer for this type, calculated as in Section 5, is more appropriate.

Situations may also arise where one is interested only in a particular class or classes of viruses found in the sample. In this case, simultaneous confidence intervals may be calcu-lated based on the number of virus types belonging to these classes, ignoring any other virus types which may be found; in addition, a single confidence interval may be calculated for the titer attributable to that class alone. One may also decide to use an "all other" category, say viruses which represent less than a given percent-age of the total, and base the analysis on grouping the all others as if they were a single type.

7. Simultaneous Confidence Intervals for Relative Fre-quencies of Virus Types

7.1 Introduction

7.1.1 Dividing the titer for a given type of virus (see Section 6) by the total PFU titer yields an estimate of the relative frequency (proportion) of that particular virus type in the sample. Alternatively, the same estimate may be obtained simply by dividing the total number of plaques found into the number of plaques testing positive for the given serotype.

7.1.2 Simultaneous 95% confidence intervals for the relative frequencies for all virus types that are identified in the sample are calculated in such a way that, as in the case of titer estimates by type of virus, the probability that all the intervals contain the true value of their respective population parameters is 95%.

7.1.3 Section 7 presents methods for obtaining 95% simultaneous confidence intervals for these proportions.

7.2 Procedure

7.2.1 Arbitrarily number the various types of viruses identified in the sample from 1 to k. Let X_h represent the number of plaques identified as being type h, h=1,2 • • • k. The proportion of virus type h is given by $p_h = X_h/X$, where X is the total number of plaques observed in all cell culture bottles.

7.2.2 If only two types of viruses have been identified in the sample (k=2), only one confidence interval need be calculated; the lower (upper) 95% confidence limit for the one type corresponds to one minus the upper (lower) 95% confidence limit for the other type. When a total of 50 or fewer viruses have been identified ($X \leq 50$), use Table 13-14 to find the lower and upper 95% confidence limits; this table is based on the exact probabilities associated with a binomial occurrence. In cases where more than 50 viruses have been typed and identified, or for results not found in Table 13-14, the 95% confidence interval for either type can be calculated using the following equation (Cochran, 1977: pp 57-59):

$$P_h \pm \left[1.96 \cdot \sqrt{\frac{P_h \, q_h}{X-1}} + \frac{1}{2X} \right]$$

where $q_h = 1 - p_h$.

7.2.3 When three or more types of viruses have been identified in the sample, the following formula may be used to determine simultaneous 95% confidence limits for the respective proportions of each type (Quesenberry and Hurst, 1964):

$$p_h^- = \frac{X^2 + 2X_h - C}{2\,(X + \chi^2)}$$

$$p_h^+ = \frac{X^2 + 2X_h + C}{2\,(X + \chi^2)}$$

where

p_h^- and p_h^+ represent the lower and upper confidence limits, respectively,

X is the total number of plaques enumerated in the sample,

X_h is the number of plaques identified as virus type h,

C is calculated as

$$C = \sqrt{\frac{\chi^2 \cdot [\chi^2 + 4 \cdot X_h(X - X_h)]}{X}}$$

The value for χ^2 (chi-square) will depend on the number of different types of viruses identified in the sample:

Number of Virus Types	χ^2
k = 3	5.731
4	6.239
5	6.635
6	6.960
7	7.237
8	7.477

The formulas for p_h^- and p_h^+ are valid as long as the lower confidence limit (p) is large enough to account for at least five viruses, given the total number of viruses identified (that is, $X \cdot p \geq 5$).

7.2.4 Table 13-15 gives selected results for this formula for the lower and upper simultaneous 95% confidence limits for the respective proportions of each type. In cases where the formula in Section 7.2.3, above, is not valid ($X \cdot p_h < 5$), the entries in Table 13-15 are based on exact multinomial probabilities, using the Bonferroni adjustment described in Section 6.4.1.

To use Table 13-15, first find the appropriate subtable based on the number of different virus types identified in the sample (k). The lower (p_h^-) and upper (p_h^+) confidence limits are found in the entries corresponding to the respective sample proportion of the virus type (p_h) and the total number of plaques found (X). Interpolation may be necessary where these values differ from the tabulated values. Table 13-15 includes only sample proportions up to 0.5; in cases where the proportion exceeds 0.5 (p_h > 0.5), simply use the table to find confidence limits for q_h, as defined in Section 7.2.2.

7.3 Examples

7.3.1 95% simultaneous confidence intervals for titer by type of virus were calculated in Section 6.3 for Dataset I. Identification of the plaques observed in this dataset are reproduced below:

Type	Number found
Coxsackievirus (CB2)	67
Coxsackievirus (CB5)	37
Poliovirus (PV1)	17

A total of 121 plaques were enumerated in all.

7.3.2 Because three types of viruses were found in the sample, the appropriate value of χ^2 to be used is 5.731. Detailed calculations for the CB2 virus are shown below:

$$C = \sqrt{\frac{5.731 \cdot [5.731 + 4 \cdot 67 \cdot (121 - 67)]}{121}}$$

$$\sqrt{685.718} = 26.186$$

$$p_1^- = \frac{5.731 + 2 \cdot 67 - 26.186}{2 \cdot (121 + 5.731)}$$

$$= \frac{113.545}{253.462} = 0.45$$

$$p_1^+ = \frac{5.731 + 2 \cdot 67 + 26.186}{2 \cdot (121 + 5.731)}$$

$$= \frac{165.917}{253.462} = 0.66$$

Calculations for all virus types found in the sample yield the following results:

Type	X_h	Sample Proportion	95% Simultaneous Confidence Interval
CB2	67	0.554	0.45 - 0.66
CB5	37	0.306	0.22 - 0.41
PV1	17	0.140	0.08 - 0.23

7.4 Rationale

7.4.1 The formula for confidence limits when two viruses have been identified is based on the normal approximation to a binomial distribution. The binomial distribution describes the probability of obtaining a given number of "successes" (one particular type of virus) out of a fixed number of "trials" (total plaques). The lower confidence limit for the true proportion of the given type of virus is such that the probability of obtaining as many or more plaques from that type of virus as were actually identified in the sample is 0.025 (2.5%). Conversely, the upper limit is such that the probability of obtaining as few or fewer plaques of that type as were identified is also 0.025. In other words, the lower (upper) confidence limit represents a value for the true relative frequency of that particular virus in the sample for which the proportion that was actually observed still may be regarded as being a "reasonable" result.

7.4.2 The term "1/(2X)" in the expression for the normal approximation to binomial confidence limits, as given in Section 7.2.2, is a *continuity correction factor*. It arises from the fact that only discrete values of p_h are possible, in increments of 1/X, while the use of the normal approximation requires that the p_h be continuously variable. More familiar may be the expression "pq/X" under the radical, rather than "pq/(X-1)". The latter term is always correct as an unbiased estimator of the sampling variance of p, although the difference between the two is not appreciable for large values of X.

7.4.3 For cases when three or more virus types are identified in the sample, the formula given in Section 7.2.3 is an approximation to the multinomial probability of finding a given number of viruses of each type. The multinomial probability distribution was used in Section 3 to test for outliers in plaque counts among cell culture bottles, where it was assumed that the probabilities of plaque formation are equal among cell culture bottles. Here, the probabilities, p_h, of finding the various types of viruses are not necessarily equal; the procedure of Section 7.2.3 establishes limits on these joint probabilities such that the observed set of proportions may be considered a likely result.

A discussion of this procedure may be found in Goodman (1965) or Quesenberry and Hurst (1964).

7.5 Implications for virus assays

7.5.1 While the procedure given in Section 6 for estimating the titer for each type of virus requires that all plaques found in each cell culture bottle, or at least a subset of these bottles, be identified, relative frequencies among the virus types can be estimated from identification of a subsample of the plaques found in each cell culture bottle. However, subsampling of plaques within a cell culture bottle is not recommended, because of the possibility that the operator's selection of plaques to be serotyped may result in substantial bias in favor of certain types of virus. If it is not feasible to identify all the plaques observed in all cell culture bottles, then a subsample of the bottles should be selected and all plaques in those bottles identified. In order to avoid bias as a result of the selection of cell culture bottles for serotyping, a fixed selection scheme should be used, such as selecting consecutively numbered bottles starting with the lowest number. Note that there is no requirement to randomize the selection of bottles, as long as the selection scheme is fixed in advance, or at least without knowledge of the results in the individual bottles.

7.5.2 As in the case of simultaneous confidence intervals for PFU titer by virus type (Section 6), simultaneous confidence intervals for proportions may depend on which virus types are considered relevant to the assay. If only viruses belonging to a particular class are of interest, then other classes of viruses may be grouped together in an "all other" category. Say that two virus types belonging to the class of interest were identified; in this case, the number of types of viruses is effectively 3 (k=3), assuming that other types of viruses were also found in the sample. One may also group viruses which represent less than a given percentage of the sample together into an "all other" category.

8. Experimental Design

8.1 Precision vs. sample size

8.1.1 Precision of the virus plaque counting assay is dependent on the total number of plaques ultimately enumerated by the assay. Assuming the PFU be randomly dispersed, the effect of total count on assay precision may be illustrated by referring to Table 13-12 for the 95% confidence limits applicable to various plaque counts. For example, say that only 10 plaques have been observed in all cell culture bottles. Table 13-12 indicates a 95% confidence interval of 4.795 to 18.39 for the true population plaque count when 10 plaques are observed in the sample; therefore, the true count may be as much as 5.205 less than or 8.39 more than the observed count. The "absolute precision" will be defined as the mean of these two extremes, which in this case is (5.205+8.39)/2 = 6.80. Notice that this corresponds to one-half the width of the 95% confidence interval. When 100 plaques are enumerated in all cell culture bottles, the corresponding absolute precision is seen to be 20.1, once again taking the half-width of the 95% confidence interval (81.36 -121.6). Absolute precision for total plaque count vs. observed plaque count is shown in Figure 13-2, and is seen to increase with total plaque count, but at a decreasing rate as the count increases.

8.1.2 Relative precision is defined as absolute precision divided by the observed count. From Section 8.1.1, above, the relative precision is 68% when 10 PFU have been enumerated (6.80/10). When 100 PFU are enumerated in all cell culture bottles, the corresponding relative precision is seen to be 20% (20.1/100). Figure 13-3 is a graph of the relative precision as defined above (half-width of the 95% confidence interval divided by observed count) vs. the observed count. Dramatic reductions in relative precision are seen to occur initially as the total count increases. Roughly, the relative precision drops by a factor of about 3/10 for a doubling of the total count; for example, relative precision is 100% for a count of 5, 68% for 10, 47% for 20, etc. Increases in the total plaque count are realized simply by assaying a larger volume of the original sample.

8.2 Sample size determination

8.2.1 The question of how much original sample needs to be assayed can be answered in terms of the degree of absolute or relative precision one desires to achieve. One problem with this approach is that one needs to know how many PFUs will be recovered from a given volume of sample in order to gauge precision; this implies that the researcher needs to know in advance what the results of the assay will be before deciding how much sample to assay in the first place, a common dilemma for sample design problems in general.

8.2.2 In many cases, a reasonable estimate of the expected PFU recovery per unit volume can be made. This is especially true if the sample is from a source that has previously been assayed for viruses; in this instance, one simply uses the last result or some average result over the last n periods for a previously sampled site and assume that the current outcome will be the same. Even when there is no historical data for a particular site, the nature of the site may enable one to make reasonable assumptions about the expected magnitude of the PFU density based on experience in assaying similar types of samples. Given an assumed value for the number of PFUs that will be recovered per unit volume of sample, or even a range of possible values, one can use Figures 13-2 and 13-3 and Table 13-12 to arrive at an adequate sample volume which will yield the desired degree of precision.

8.2.3 Another solution to sample size determination is to perform a screening experiment on the sample. When historical data from the sampling site is lacking, a screening experiment is also desirable from the standpoint of determining proper dilution levels for the eluate; this in itself is important to maximizing the precision of the assay by avoiding loss of data through cases where plaques are too numerous to count (TNTC). However, in many instances, the extra time and expense involved in performing a screening experiment may be prohibitive.

8.2.4 A practical approach to determining the total volume of sample to be assayed is to establish some minimum *absolute* precision level when the PFU density in the original sample falls below some predetermined low level while targeting for a minimum *relative* precision for densities beyond some other, higher value of PFU density. For example, one may set the estimation error to be no more than 20% (relative precision) if the PFU density in the original sample is at least 50 PFU/L, but be willing to accept estimation error of 5 PFU/L (absolute precision) if the PFU density is found to be 10 PFU/L or less. These two criteria lead to two sample size requirements:

(a) For a relative precision of 20%, about 100 total plaques need to be enumerated in the assay (Figure 13-3). When the density of PFUs in the sample is 50 per liter, two liters of sample need to be assayed to achieve a total plaque count of 100. If the density is higher than 50 PFU/L, then assaying two liters of sample will yield a plaque count over 100, and, as can be seen in Figure 13-3, the relative precision will be better than 20%.

(b) An absolute precision of 5 PFU/L corresponds to a relative precision of 50% when the true density is 10 PFU/mL; from Table 13-12, the relative precision is 49.3% when the sample plaque count totals 18. Therefore, with a density of 10 PFU/L, 1.8 liters of sample are required in order to achieve the second objective. If the density is less than 10 PFU/L, then fewer than 18 plaques are expected to be enumerated, and, as can be seen from Figure 13-2, the absolute sampling error will be lower than 5 PFU/L (absolute precision in total plaque count < 9).

(c) The larger of the two sample requirements, in this case two liters, is used. The stricter requirement turns out to be the limit of 20% relative precision for a density of 50 PFU/L or more.

8.2.5 When adsorption, elution, and/or concentration methods are used in recovering viruses, choices must be made as to the volume of sample to be used in the recovery procedure, whether to further concentrate the eluate, the volume and dilutions of eluate to be inoculated, and the number of cell culture bottles to receive these inoculations. As long as the number of plaques ultimately enumerated in the assay are sufficient to meet the sampling requirements, as described above, and assuming that PFUs are randomly dispersed, these details of the experimental design do not affect sampling precision, and are not relevant to sample size considerations.

8.2.6 Other factors, such as cost or time, may be considered in making these decisions. As an illustration, consider the sample size requirement derived in the previous section. Assume that the adsorption-elution process is expected to yield about 40 mL of eluate, and that a 1:10 dilution series will be used so that cell cultures will be inoculated with 1, 0.1, and 0.01 mL of eluate. Three cell culture bottles, one at each dilution level, will be considered as comprising a single replicate; therefore, one replicate accounts for 1.11 mL of eluate volume. The following represent different ways in which two liters of sample can be subjected to the assay in terms of the initial sample volume used in the adsorption-elution procedure and the number of replicate observations:

Initial Sample Volume (L)	Replicates	Cell Culture Bottles	Total Eluate Volume Used (mL)
2	36	108	39.96
4	18	54	19.98
8	9	27	9.99

In general, it is desirable to use a dilution series, similar to that in this example, and to use as many replicates as feasible in order to guard against potential data losses due to TNTC situations, toxicity, or other effects which may lead to the loss of results in individual cell culture bottles.

8.3 Sampling requirements for estimation by virus type

8.3.1 The procedure given in Section 6 for estimating PFU titer by type of virus is valid only when all plaques found in a set of cell culture bottles, if not all cell culture bottles, are serotyped. The method of Section 7 for estimating relative frequencies of virus types, however, can be applied even if only selected plaques from each cell culture bottle are typed, with the minor modification that the total number of plaques enumerated be replaced by the total number of plaques which have been so identified. Nonetheless, even if only relative frequency, and not titer by virus type were of interest, the recommendation is to identify all of the plaques found in a cell culture bottle until all plaques enumerated in the assay have been identified, or until a number of plaques have been identified which is sufficient to meet precision requirements. This eliminates bias that may result from operator selection of specific plaques to be identified.

8.3.2 Estimating sample size requirements for attaining a target precision for PFU density or frequency by type of virus is more uncertain than estimating sample size requirements based on overall PFU density; precision now depends on the number of different types one expects to find and their expected relative

frequencies, in addition to the total PFU density. Generally, unless it is very important to obtain precise estimates of densities of the various types of viruses that may be found in the sample, one designs the assay for total PFU recovery and accepts whatever result occurs with respect to virus type.

9. References

American Society for Testing and Materials: Committee E-11 on Statistical Methods. 1980. Standard practice for dealing with outlying observations, ASTM E 178-180.

Barnett, V. and Lewis, T. 1984. Outliers in Statistical Data, 2nd ed., John Wiley and Sons, NY.

Cochran, W.G. 1954. Some methods for strengthening the common χ^2 tests. *Biometrics* 10:417-451.

Cochran, W.G. 1977. Sampling Techniques, 3rd ed., John Wiley and Sons, NY.

Cox, D.R. 1953. Some simple approximate tests for Poisson variates. *Biometrika* 40:354-360.

Garwood, F. 1936. Fiducial limits for the Poisson distribution. *Biometrika* 43:423-435.

Fisher, R.A. 1950. The significance of deviations from expectations in a Poisson series. *Biometrics* 6:17-24.

Goodman, L.A. 1965. On simultaneous confidence intervals for multinomial proportions. *Technometrics* 7:247-254.

Quesenberry, C.P. and Hurst, D.C. 1964. Large sample simultaneous confidence intervals for multinomial proportions. *Technometrics* 6:191-195.

Sachs, L. 1984. Applied Statistics: A Handbook of Techniques, 2nd ed., Springer-Verlag, NY.

Snedecor, G.W. and Cochran, W.G. 1980. Statistical Methods, 7th ed., Iowa State University Press, Ames, Iowa.

Table 13-1. *Test of Dispersion for Dataset I*

(1) i	(2) bottle number(s)	(3) v_i (mL)	(4) Expected count ($t \cdot v_i$)	(5) Observed count xi	(6) $\frac{(x_i - t \cdot v_i)^2}{t \cdot v_i}$
1	1-10	.01	1.09	0	1.090
2	11-15	.05	5.45	6	0.056
3	16-20	.05	5.45	2	2.184
4	21	.1	10.9	10	0.074
5	22	.1	10.9	12	0.111
6	23	.1	10.9	10	0.074
7	24	.1	10.9	6	2.203
8	25	.1	10.9	16	2.386
9	26	.1	10.9	13	0.405
10	27	.1	10.9	9	0.331
11	28	.1	10.9	14	0.882
12	29	.1	10.9	6	2.203
13	30	.1	10.9	17	3.414
TOTAL		1.11	121	121	15.413

$$D = 15.413 \; < \chi^2_{12,.05} = 21.026$$

Table 13-2. *Test of Dispersion for Dataset II*

(1) i	(2) bottle number	(3) v_i (mL)	(4) Expected count ($\bar{x}$)	(5) Observed count x_i	(6) $(x_i - \bar{x})^2 / \bar{x}$
1	1	1	11.857	7	1.990
2	2	1	11.857	14	0.387
3	3	1	11.857	7	1.990
4	4	1	11.857	10	0.291
5	5	1	11.857	15	0.833
6	6	1	11.857	21	7.050
7	7	1	11.857	9	0.688
8	8	1	11.857	18	3.182
9	9	1	11.857	6	2.893
10	10	1	11.857	6	2.893
11	11	1	11.857	20	5.592
12	12	1	11.857	8	1.255
13	13	1	11.857	17	2.231
14	14	1	11.857	8	1.255
TOTAL		14	166	166	32.530

$$D = 32.530 \; < \chi^2_{13,.05} = 22.362$$

Table 13-3. *0.05 Critical Values of the Chi-Square (χ^2) Cumulative Distribution*

Degrees of freedom	Chi* square	Degrees of freedom	Chi square
1	3.841	26	38.885
2	5.991	27	40.113
3	7.815	28	41.337
4	9.488	29	42.557
5	11.070	30	43.773
6	12.592	31	44.985
7	14.067	32	46.194
8	15.507	33	47.400
9	16.919	34	48.602
10	18.307	35	49.802
11	19.675	36	50.998
12	21.026	37	52.192
13	22.362	38	53.384
14	23.685	39	54.572
15	24.996	40	55.758
16	26.296	41	56.942
17	27.587	42	58.124
18	28.869	43	59.304
19	30.144	44	60.481
20	31.410	45	61.656
21	32.671	46	62.830
22	33.924	47	64.001
23	35.172	48	65.171
24	36.415	49	66.339
25	37.652	50	67.505

*For degrees of freedom (v) > 50, use $\chi^2 = 1/2 \bullet (1.645 + \sqrt{2\nu-1})^2$

Table 13-4. *0.01 Critical Values for a Single Upper Outlier from a Poisson (Random) Distribution*

Highest count	Number of cell culture bottles: 3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20
Is an upper outlier when the total count is less than																		
3	-	-	-	-	-	-	-	4	4	4	4	4	4	4	4	4	4	5
4	-	-	5	5	5	6	6	6	7	7	7	7	8	8	8	9	9	9
5	-	6	7	7	8	9	9	10	10	11	11	12	12	13	13	14	14	15
6	7	8	9	10	11	12	13	14	15	15	16	17	18	19	20	20	21	22
7	9	10	12	13	14	16	17	18	19	21	22	23	24	25	26	28	29	30
8	10	12	14	16	18	20	21	23	25	26	28	29	31	32	34	36	37	39
9	12	15	17	19	22	24	26	28	30	32	34	36	38	40	42	44	46	48
10	14	17	20	23	25	28	31	33	36	38	41	43	46	48	51	53	55	58
11	16	20	23	26	29	33	36	39	42	45	48	51	54	57	60	62	65	68
12	18	22	26	30	34	37	41	45	48	52	55	59	62	65	69	72	75	79
13	20	25	29	34	38	42	46	50	55	59	63	67	71	74	78	82	86	90
14	22	28	33	38	42	47	52	57	61	66	70	75	79	84	88	93	97	101
15	25	30	36	41	47	52	58	63	68	73	78	83	88	93	98	103	108	113
16	27	33	39	45	51	57	63	69	75	81	86	92	97	103	109	114	120	125
17	29	36	43	49	56	63	69	75	82	88	94	101	107	113	119	125	131	137
18	31	39	46	54	61	68	75	82	89	96	103	110	116	123	130	136	143	150
19	33	42	50	58	66	73	81	89	96	104	111	119	126	133	141	148	155	162
20	36	44	53	62	70	79	87	95	104	112	120	128	136	144	152	159	167	175
21	38	47	57	66	75	84	93	102	111	120	128	137	146	154	163	171	180	188
22	40	50	60	70	80	90	99	109	118	128	137	146	156	165	174	183	192	201
23	42	53	64	75	85	95	106	116	126	136	146	156	166	176	185	195	205	214
24	45	56	68	79	90	101	112	123	134	144	155	166	176	186	197	207	218	228
25	47	59	71	83	95	107	118	130	141	153	164	175	186	197	209	220	231	241
26	49	62	75	88	100	113	125	137	149	161	173	185	197	209	220	232	244	255
27	52	65	79	92	105	118	131	144	157	170	182	195	207	220	232	244	257	269
28	54	69	83	97	111	124	138	151	165	178	192	205	218	231	244	257	270	283
29	57	72	87	101	116	130	145	159	173	187	201	215	229	242	256	270	283	297
30	59	75	90	106	121	136	151	166	181	196	210	225	239	254	268	282	297	311
31	61	78	94	110	126	142	158	173	189	204	220	235	250	265	280	295	310	325
32	64	81	98	115	132	148	164	181	197	213	229	245	261	277	293	308	324	340
33	66	84	102	119	137	154	171	188	205	222	239	255	272	288	305	321	338	354
34	69	87	106	124	142	160	178	196	213	231	248	266	283	300	317	334	351	368
35	71	90	110	129	148	166	185	203	222	240	258	276	294	312	330	348	365	383
36	73	94	114	133	153	172	192	211	230	249	268	286	305	324	342	361	379	398
37	76	97	118	138	158	178	198	218	238	258	277	297	316	336	355	374	393	412
38	78	100	122	143	164	185	205	226	246	267	287	307	327	348	368	387	407	427
39	81	103	125	147	169	191	212	234	255	276	297	318	339	360	380	401	422	442
40	83	107	129	152	175	197	219	241	263	285	307	329	350	372	393	414	436	457
41	86	110	133	157	180	203	226	249	272	294	317	339	361	384	406	428	450	472
42	88	113	137	162	186	210	233	257	280	303	327	350	373	396	419	442	464	487
43	91	116	141	166	191	216	240	264	289	313	337	361	384	408	432	455	479	502
44	93	120	145	171	197	222	247	272	297	322	347	371	396	420	445	469	493	517
45	96	123	150	176	202	228	254	280	306	331	357	382	407	432	458	483	508	532
46	98	126	154	181	208	235	261	288	314	341	367	393	419	445	471	496	522	548
47	101	129	158	186	213	241	268	296	323	350	377	404	430	457	484	510	537	563
48	103	133	162	190	219	247	276	304	332	359	387	415	442	470	497	524	551	578
49	106	136	166	195	225	254	283	312	340	369	397	426	454	482	510	538	566	594
50	108	139	170	200	230	260	290	319	349	378	407	436	465	494	523	552	581	609

(continued)

Table 13-4. (continued)

Highest count is an upper outlier when the total count is less than	Number of cell culture bottles																	
	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20
51	111	143	174	205	236	267	297	327	358	388	418	447	477	507	536	566	595	625
52	113	146	178	210	242	273	304	335	366	397	428	458	489	519	550	580	610	640
53	116	149	182	215	247	279	311	343	375	407	438	470	501	532	563	594	625	656
54	118	153	186	220	253	286	319	351	384	416	448	481	513	545	576	608	640	672
55	121	156	190	225	259	292	326	359	393	426	459	492	524	557	590	622	655	687
56	124	159	195	230	264	299	333	367	401	435	469	503	536	570	603	637	670	703
57	126	163	199	235	270	305	341	375	410	445	480	514	548	583	617	651	685	719
58	129	166	203	239	276	312	348	384	419	455	490	525	560	595	630	665	700	734
59	131	169	207	244	282	318	355	392	428	464	500	536	572	608	644	679	715	750
60	134	173	211	249	287	325	362	400	437	474	511	548	584	621	657	694	730	766
61	136	176	215	254	293	331	370	408	446	484	521	559	596	634	671	708	745	782
62	139	179	220	259	299	338	377	416	455	493	532	570	608	646	684	722	760	798
63	142	183	224	264	305	345	384	424	464	503	542	581	620	659	698	737	775	814
64	144	186	228	269	310	351	392	432	473	513	553	593	632	672	712	751	791	830
65	147	190	232	274	316	358	399	440	482	522	563	604	645	685	725	766	806	846
66	149	193	236	279	322	364	407	449	491	532	574	615	657	698	739	780	821	862
67	152	196	241	284	328	371	414	457	500	542	584	627	669	711	753	795	836	878
68	155	200	245	289	334	378	421	465	509	552	595	638	681	724	767	809	852	894
69	157	203	249	294	339	384	429	473	518	562	606	649	693	737	780	824	867	910
70	160	207	253	299	345	391	436	482	527	572	616	661	705	750	794	838	882	926
71	162	210	257	304	351	398	444	490	536	581	627	672	718	763	808	853	898	943
72	165	214	262	309	357	404	451	498	545	591	638	684	730	776	822	868	913	959
73	168	217	266	315	363	411	459	506	554	601	648	695	742	789	836	882	929	975
74	170	220	270	320	369	418	466	515	563	611	659	707	754	802	850	897	944	991
75	173	224	274	325	375	424	474	523	572	621	670	718	767	815	863	912	960	1008
76	175	227	279	330	381	431	481	531	581	631	680	730	779	828	877	926	975	1024
77	178	231	283	335	386	438	489	540	590	641	691	741	791	841	891	941	991	1040
78	181	234	287	340	392	444	496	548	599	651	702	753	804	855	905	956	1006	1057
79	183	238	292	345	398	451	504	556	609	661	713	765	816	868	919	971	1022	1073
80	186	241	296	350	404	458	511	565	618	671	723	776	829	881	933	985	1037	1089
81	189	245	300	355	410	465	519	573	627	681	734	788	841	894	947	1000	1053	1106
82	191	248	304	360	416	471	527	581	636	691	745	799	853	907	961	1015	1069	1122
83	194	251	309	365	422	478	534	590	645	701	756	811	866	921	975	1030	1084	1139
84	196	255	313	371	428	485	542	598	655	711	767	823	878	934	989	1045	1100	1155
85	199	258	317	376	434	492	549	607	664	721	778	834	891	947	1004	1060	1116	1172
86	202	262	322	381	440	498	557	615	673	731	788	846	903	961	1018	1075	1132	1188
87	204	265	326	386	446	505	564	623	682	741	799	858	916	974	1032	1090	1147	1205
88	207	269	330	391	452	512	572	632	692	751	810	869	928	987	1046	1105	1163	1222
89	210	272	334	396	458	519	580	640	701	761	821	881	941	1001	1060	1120	1179	1238
90	212	276	339	401	464	526	587	649	710	771	832	893	953	1014	1074	1135	1195	1255
91	215	279	343	407	470	532	595	657	719	781	843	905	966	1027	1088	1150	1210	1271
92	218	283	347	412	476	539	603	666	729	791	854	916	979	1041	1103	1165	1226	1288
93	220	286	352	417	482	546	610	674	738	801	865	928	991	1054	1117	1180	1242	1305
94	223	290	356	422	488	553	618	683	747	812	876	940	1004	1068	1131	1195	1258	1321
95	226	293	360	427	494	560	626	691	757	822	887	952	1016	1081	1145	1210	1274	1338
96	228	297	365	432	500	567	633	700	766	832	898	963	1029	1094	1160	1225	1290	1355
97	231	300	369	438	506	573	641	708	775	842	909	975	1042	1108	1174	1240	1306	1372
98	234	304	373	443	512	580	649	717	785	852	920	987	1054	1121	1188	1255	1322	1388
99	236	307	378	448	518	587	656	725	794	862	931	999	1067	1135	1203	1270	1338	1405
100	239	311	382	453	524	594	664	734	803	873	942	1011	1080	1148	1217	1285	1354	1422

Table 13-5. *0.01 Critical Values for a Single Lower Outlier from a Poisson (Random) Distribution*

Lowest count Is a lower outlier when the total count is greater than	Number of cell culture bottles: 3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20
0	14	20	27	35	42	50	57	65	73	81	89	97	105	114	122	131	139	148
1	19	29	38	48	57	67	77	88	98	108	119	130	140	151	162	173	184	195
2	25	36	47	59	71	83	95	107	119	132	145	157	170	183	196	209	222	235
3	29	42	56	69	83	97	111	125	139	154	168	183	198	212	227	242	257	273
4	34	49	64	79	94	110	126	142	158	174	191	207	224	240	257	274	291	308
5	38	55	71	88	106	123	140	158	176	194	212	230	249	267	285	304	323	341
6	43	61	79	98	116	135	155	174	193	213	233	253	273	293	313	333	353	374
7	47	66	86	106	127	147	168	189	210	232	253	275	296	318	340	362	384	406
8	51	72	94	115	137	159	182	204	227	250	273	296	319	342	366	389	413	437
9	55	78	101	124	147	171	195	219	243	268	292	317	342	367	391	417	442	467
10	59	83	108	132	157	183	208	234	259	285	311	337	364	390	417	443	470	497
11	63	89	114	141	167	194	221	248	275	303	330	358	386	414	442	470	498	526
12	67	94	121	149	177	205	233	262	291	320	349	378	407	437	466	496	525	555
13	71	99	128	157	187	216	246	276	306	337	367	398	429	459	490	521	553	584
14	75	105	135	165	196	227	258	290	321	353	385	417	450	482	514	547	580	612
15	79	110	141	173	205	238	271	304	337	370	403	437	470	504	538	572	606	640
16	83	115	148	181	215	249	283	317	352	386	421	456	491	526	562	597	633	668
17	86	120	155	189	224	259	295	331	367	403	439	475	512	548	585	622	659	696
18	90	125	161	197	233	270	307	344	381	419	456	494	532	570	608	647	685	723
19	94	130	167	205	243	281	319	357	396	435	474	513	552	592	631	671	711	751
20	98	136	174	213	252	291	331	371	411	451	491	532	573	613	654	695	736	778
21	101	141	180	220	261	302	343	384	425	467	509	550	593	635	677	719	762	805
22	105	146	187	228	270	312	354	397	440	483	526	569	612	656	700	743	787	831
23	109	151	193	236	279	322	366	410	454	498	543	587	632	677	722	767	813	858
24	112	156	199	243	288	333	378	423	468	514	560	606	652	698	745	791	838	884
25	116	161	206	251	297	343	389	436	483	530	577	624	671	719	767	815	863	911
26	120	166	212	259	306	353	401	449	497	545	594	642	691	740	789	838	888	937
27	123	170	218	266	314	363	412	461	511	560	610	660	710	761	811	862	912	963
28	127	175	224	274	323	373	424	474	525	576	627	678	730	781	833	885	937	989
29	131	180	230	281	332	383	435	487	539	591	644	696	749	802	855	908	962	1015
30	134	185	237	288	341	393	446	499	553	606	660	714	768	822	877	931	986	1041
31	138	190	243	296	349	403	458	512	567	622	677	732	787	843	899	954	1010	1066
32	141	195	249	303	358	413	469	525	581	637	693	750	806	863	920	977	1035	1092
33	145	200	255	311	367	423	480	537	594	652	710	767	825	884	942	1000	1059	1118
34	149	205	261	318	376	433	491	550	608	667	726	785	844	904	963	1023	1083	1143
35	152	209	267	325	384	443	503	562	622	682	742	803	863	924	985	1046	1107	1168
36	156	214	273	333	393	453	514	575	636	697	759	820	882	944	1006	1069	1131	1194
37	159	219	279	340	401	463	525	587	649	712	775	838	901	964	1028	1091	1155	1219
38	163	224	285	347	410	473	536	599	663	727	791	855	920	984	1049	1114	1179	1244
39	166	228	291	355	418	482	547	612	677	742	807	873	938	1004	1070	1136	1203	1269
40	170	233	297	362	427	492	558	624	690	757	823	890	957	1024	1091	1159	1227	1294
41	173	238	303	369	435	502	569	636	704	771	839	907	976	1044	1113	1181	1250	1319
42	177	243	309	376	444	512	580	648	717	786	855	925	994	1064	1134	1204	1274	1344
43	180	247	315	384	452	521	591	661	731	801	871	942	1013	1084	1155	1226	1297	1369
44	184	252	321	391	461	531	602	673	744	816	887	959	1031	1103	1176	1248	1321	1394
45	187	257	327	398	469	541	613	685	758	830	903	976	1050	1123	1197	1271	1344	1418
46	191	262	333	405	478	551	624	697	771	845	919	993	1068	1143	1218	1293	1368	1443
47	194	266	339	412	486	560	635	709	784	859	935	1011	1086	1162	1239	1315	1391	1468
48	198	271	345	419	494	570	645	721	798	874	951	1028	1105	1182	1259	1337	1415	1492
49	201	276	351	427	503	579	656	733	811	889	967	1045	1123	1202	1280	1359	1438	1517
50	205	280	357	434	511	589	667	746	824	903	982	1062	1141	1221	1301	1381	1461	1541

Table 13-6. *0.01 Critical Values for a Pair of Upper Outlier from a Poisson (Random) Distribution*

Sum of 2 Highest counts	Number of cell culture bottles: 5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20
	Are an upper outlier pair when the total count is less than															
4	-	-	-	-	-	-	-	5	5	5	5	5	5	5	5	5
5	-	-	6	6	6	6	6	7	7	7	7	7	7	8	8	8
6	7	7	7	8	8	8	9	9	9	10	10	10	11	11	11	11
7	8	9	9	10	10	11	11	12	12	13	13	14	14	14	15	15
8	10	11	11	12	13	13	14	15	15	16	17	17	18	18	19	19
9	11	12	13	14	15	16	17	18	19	19	20	21	22	23	23	24
10	13	14	15	17	18	19	20	21	22	23	24	25	26	27	28	29
11	15	16	18	19	20	22	23	24	26	27	28	29	30	32	33	34
12	17	18	20	22	23	25	26	28	29	31	32	34	35	36	38	39
13	18	20	22	24	26	28	30	31	33	35	37	38	40	41	43	45
14	20	22	25	27	29	31	33	35	37	39	41	43	45	47	48	50
15	22	25	27	30	32	34	37	39	41	43	45	48	50	52	54	56
16	24	27	30	32	35	38	40	43	45	48	50	53	55	57	60	62
17	26	29	32	35	38	41	44	47	49	52	55	57	60	63	65	68
18	28	31	35	38	41	44	47	51	54	57	60	63	65	68	71	74
19	30	33	37	41	44	48	51	55	58	61	64	68	71	74	77	80
20	32	36	40	44	47	51	55	59	62	66	69	73	76	80	83	87
21	33	38	42	46	51	55	59	63	67	70	74	78	82	86	89	93
22	35	40	45	49	54	58	63	67	71	75	79	83	88	92	96	100
23	37	42	47	52	57	62	66	71	76	80	84	89	93	98	102	106
24	39	45	50	55	60	65	70	75	80	85	90	94	99	104	108	113
25	41	47	53	58	64	69	74	79	85	90	95	100	105	110	115	119
26	43	49	55	61	67	73	78	84	89	95	100	105	111	116	121	126
27	45	52	58	64	70	76	82	88	94	100	105	111	117	122	128	133
28	47	54	61	67	74	80	86	92	99	105	111	117	122	128	134	140
29	49	57	64	70	77	84	90	97	103	110	116	122	128	135	141	147
30	52	59	66	73	81	88	94	101	108	115	121	128	135	141	147	154
31	54	61	69	77	84	91	99	106	113	120	127	134	141	147	154	161
32	56	64	72	80	87	95	103	110	118	125	132	140	147	154	161	168
33	58	66	75	83	91	99	107	115	122	130	138	145	153	160	168	175
34	60	69	77	86	94	103	111	119	127	135	143	151	159	167	175	182
35	62	71	80	89	98	107	115	124	132	141	149	157	165	173	182	190
36	64	74	83	92	101	110	119	128	137	146	154	163	172	180	188	197
37	66	76	86	95	105	114	124	133	142	151	160	169	178	187	195	204
38	68	78	89	99	108	118	128	137	147	156	166	175	184	193	202	212
39	70	81	91	102	112	122	132	142	152	162	171	181	190	200	209	219
40	72	83	94	105	116	126	136	147	157	167	177	187	197	207	217	226
41	74	86	97	108	119	130	141	151	162	172	183	193	203	213	224	234
42	77	88	100	111	123	134	145	156	167	178	188	199	210	220	231	241
43	79	91	103	115	126	138	149	161	172	183	194	205	216	227	238	249
44	81	93	106	118	130	142	154	165	177	188	200	211	223	234	245	256
45	83	96	109	121	133	146	158	170	182	194	206	217	229	241	252	264
46	85	98	111	124	137	150	162	175	187	199	211	224	236	248	259	271
47	87	101	114	128	141	154	167	179	192	205	217	230	242	254	267	279
48	89	103	117	131	144	158	171	184	197	210	223	236	249	261	274	287
49	91	106	120	134	148	162	175	189	202	216	229	242	255	268	281	294
50	94	108	123	137	152	166	180	194	208	221	235	248	262	275	289	302

(continued)

Table 13-6. *(continued)*

Sum of 2 Highest counts	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20
	Number of cell culture bottles — Are an upper outlier pair when the total count is less than															
51	96	111	126	141	155	170	184	199	213	227	241	255	268	282	296	310
52	98	113	129	144	159	174	189	203	218	232	247	261	275	289	303	317
53	100	116	132	147	163	178	193	208	223	238	253	267	282	296	311	325
54	102	119	135	151	166	182	198	213	228	243	258	273	288	303	318	333
55	104	121	138	154	170	186	202	218	233	249	264	280	295	310	325	341
56	107	124	141	157	174	190	206	223	239	255	270	286	302	317	333	348
57	109	126	144	161	178	194	211	227	244	260	276	292	309	324	340	356
58	111	129	146	164	181	198	215	232	249	266	282	299	315	332	348	364
59	113	131	149	167	185	203	220	237	254	271	288	305	322	339	355	372
60	115	134	152	171	189	207	224	242	260	277	294	312	329	346	363	380
61	117	137	155	174	192	211	229	247	265	283	300	318	336	353	370	388
62	120	139	158	177	196	215	233	252	270	288	306	324	342	360	378	396
63	122	142	161	181	200	219	238	257	275	294	312	331	349	367	385	404
64	124	144	164	184	204	223	242	262	281	300	319	337	356	375	393	411
65	126	147	167	187	207	227	247	267	286	305	325	344	363	382	401	419
66	128	149	170	191	211	231	252	272	291	311	331	350	370	389	408	427
67	131	152	173	194	215	236	256	276	297	317	337	357	376	396	416	435
68	133	155	176	198	219	240	261	281	302	322	343	363	383	403	423	443
69	135	157	179	201	223	244	265	286	307	328	349	370	390	411	431	451
70	137	160	182	204	226	248	270	291	313	334	355	376	397	418	439	459
71	139	162	185	208	230	252	274	296	318	340	361	383	404	425	446	468
72	142	165	188	211	234	257	279	301	323	345	367	389	411	433	454	476
73	144	168	191	215	238	261	284	306	329	351	374	396	418	440	462	484
74	146	170	194	218	242	265	288	311	334	357	380	402	425	447	470	492
75	148	173	197	221	245	269	293	316	340	363	386	409	432	455	477	500

(continued)

Table 13-6. (continued)

Sum of 2 Highest counts	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20
	Number of cell culture bottles — Are an upper outlier pair when the total count is less than															
76	150	176	200	225	249	273	297	321	345	369	392	415	439	462	485	508
77	153	178	203	228	253	278	302	326	350	374	398	422	446	469	493	516
78	155	181	206	232	257	282	307	331	356	380	404	429	453	477	500	524
79	157	183	209	235	261	286	311	336	361	386	411	435	460	484	508	532
80	159	186	212	239	265	290	316	341	367	392	417	442	467	491	516	541
81	161	189	215	242	268	295	321	346	372	398	423	448	474	499	524	549
82	164	191	219	245	272	299	325	351	377	403	429	455	481	506	532	557
83	166	194	222	249	276	303	330	356	383	409	435	462	488	514	539	565
84	168	197	225	252	280	307	334	361	388	415	442	468	495	521	547	573
85	170	199	228	256	284	312	339	367	394	421	448	475	502	528	555	582
86	173	202	231	259	288	316	344	372	399	427	454	482	509	536	563	590
87	175	204	234	263	292	320	348	377	405	433	461	488	516	543	571	598
88	177	207	237	266	295	324	353	382	410	439	467	495	523	551	579	606
89	179	210	240	270	299	329	358	387	416	444	473	502	530	558	586	615
90	182	212	243	273	303	333	363	392	421	450	479	508	537	566	594	623
91	184	215	246	277	307	337	367	397	427	456	486	515	544	573	602	631
92	186	218	249	280	311	341	372	402	432	462	492	522	551	581	610	639
93	188	220	252	284	315	346	377	407	438	468	498	528	558	588	618	648
94	191	223	255	287	319	350	381	412	443	474	505	535	565	596	626	656
95	193	226	258	291	323	354	386	417	449	480	511	542	573	603	634	664
96	195	228	261	294	326	359	391	423	454	486	517	548	580	611	642	673
97	197	231	264	298	330	363	395	428	460	492	524	555	587	618	650	681
98	200	234	267	301	334	367	400	433	465	498	530	562	594	626	658	689
99	202	236	271	304	338	372	405	438	471	504	536	569	601	633	666	698
100	204	239	274	308	342	376	410	443	476	510	543	575	608	641	673	706

Table 13-7. *0.01 Critical Values for a Pair of Lower Outliers from a Poisson (Random) Distribution*

Sum of 2 Lowest counts	Number of cell culture bottles: 5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20
	Are a lower outlier pair when the total count is greater than															
0	10	14	17	21	26	30	34	39	43	48	53	58	62	67	72	77
1	15	19	25	30	35	41	46	52	58	64	70	76	82	89	95	101
2	19	25	31	37	44	50	57	64	71	78	85	92	99	107	114	122
3	23	29	36	44	51	59	67	74	82	90	99	107	115	124	132	141
4	26	34	42	50	59	67	76	85	93	102	112	121	130	139	149	158
5	30	38	47	56	66	75	85	94	104	114	124	134	144	154	165	175
6	33	43	52	62	72	83	93	104	114	125	136	147	158	169	180	192
7	37	47	57	68	79	90	101	113	124	136	148	159	171	183	195	208
8	40	51	62	74	86	98	110	122	134	147	159	172	184	197	210	223
9	43	55	67	80	92	105	118	131	144	157	170	184	197	211	225	238
10	47	59	72	85	98	112	125	139	153	167	181	196	210	224	239	254
11	50	63	77	91	105	119	133	148	162	177	192	207	222	238	253	268
12	53	67	81	96	111	126	141	156	172	187	203	219	235	251	267	283
13	56	71	86	101	117	133	149	165	181	197	214	230	247	264	280	297
14	59	75	91	107	123	139	156	173	190	207	224	241	259	276	294	312
15	63	79	95	112	129	146	164	181	199	217	235	253	271	289	307	326
16	66	83	100	117	135	153	171	189	208	226	245	264	283	302	321	340
17	69	86	104	122	141	159	178	197	216	236	255	275	294	314	334	354
18	72	90	109	128	147	166	186	205	225	245	265	286	306	326	347	368
19	75	94	113	133	153	173	193	213	234	255	275	296	317	339	360	381
20	78	98	118	138	158	179	200	221	242	264	285	307	329	351	373	395
21	81	101	122	143	164	186	207	229	251	273	295	318	340	363	386	408
22	84	105	126	148	170	192	214	237	260	282	305	328	352	375	398	422
23	87	109	131	153	176	198	221	245	268	292	315	339	363	387	411	435
24	90	112	135	158	181	205	228	252	276	301	325	349	374	399	424	449
25	93	116	139	163	187	211	236	260	285	310	335	360	385	411	436	462
26	96	120	144	168	193	217	243	268	293	319	344	370	396	422	449	475
27	99	123	148	173	198	224	249	275	302	328	354	381	407	434	461	488
28	102	127	152	178	204	230	256	283	310	337	364	391	418	446	473	501
29	105	131	157	183	209	236	263	291	318	346	373	401	429	458	486	514
30	108	134	161	188	215	243	270	298	326	355	383	412	440	469	498	527
31	111	138	165	193	221	249	277	306	335	363	393	422	451	481	510	540
32	114	141	169	198	226	255	284	313	343	372	402	432	462	492	523	553
33	117	145	173	202	232	261	291	321	351	381	412	442	473	504	535	566
34	120	148	178	207	237	267	298	328	359	390	421	452	484	515	547	579
35	123	152	182	212	243	273	304	336	367	399	430	462	494	527	559	591
36	125	156	186	217	248	280	311	343	375	407	440	472	505	538	571	604
37	128	159	190	222	254	286	318	351	383	416	449	482	516	549	583	617
38	131	163	194	227	259	292	325	358	391	425	459	492	526	561	595	629
39	134	166	199	231	264	298	331	365	399	434	468	502	537	572	607	642
40	137	170	203	236	270	304	338	373	407	442	477	512	548	583	619	655
41	140	173	207	241	275	310	345	380	415	451	487	522	558	594	631	667
42	143	177	211	246	281	316	352	387	423	459	496	532	569	606	643	680
43	146	180	215	250	286	322	358	395	431	468	505	542	579	617	654	692
44	149	184	219	255	292	328	365	402	439	477	514	552	590	628	666	704
45	151	187	223	260	297	334	372	409	447	485	523	562	600	639	678	717
46	154	191	228	265	302	340	378	417	455	494	533	572	611	650	690	729
47	157	194	232	269	308	346	385	424	463	502	542	581	621	661	701	742
48	160	198	236	274	313	352	391	431	471	511	551	591	632	672	713	754
49	163	201	240	279	318	358	398	438	479	519	560	601	642	683	725	766
50	166	205	244	284	324	364	405	445	487	528	569	611	653	694	737	779

Table 13-8. 0.01 Critical Values for the Test for a Single Upper ("T_n") or Lower ("T_1") Outlier from a Normal Distribution

Number of cell culture bottles	Critical T value	Number of cell culture bottles	Critical T value
3	1.155	17	2.785
4	1.492	18	2.821
5	1.749	19	2.854
6	1.944	20	2.884
7	2.097	21	2.912
8	2.221	22	2.939
9	2.323	23	2.963
10	2.410	24	2.987
11	2.484	25	3.009

Table 13-9. 0.01 Critical Values for the Test for an Upper ("$T_{n-1,n}$") or Lower ("$T_{1,2}$") Outlier Pair from a Normal Distribution

Number of cell culture bottles	Critical T value	Number of cell culture bottles	Critical T value
4	0.0000	18	0.3530
5	0.0035	19	0.3725
6	0.0186	20	0.3909
7	0.0440	21	0.4082
8	0.0750	22	0.4245
9	0.1082	23	0.4398
10	0.1414	24	0.4543
11	0.1736	25	0.4680
12	0.2043	26	0.4810
13	0.2333	27	0.4933
14	0.2605	28	0.5050
15	0.2859	29	0.5162
16	0.3098	30	0.5268
17	0.3321		

* *Upper or lower outlier pairs are statistically significant outliers if T_{n-1},n or $T_{1,2}$ is less than the tabulated values*

Table 13-10 0.05 Two-Tailed Critical Values for the Cumulative F Distribution

	Numerator degrees of freedom														
DF*	1	2	5	10	15	20	25	30	40	50	100	200	500	1000	∞
1	648	800	922	969	985	993	998	1001	1006	1008	1013	1016	1017	1018	1018
2	38.51	39.00	39.30	39.40	39.43	39.45	39.46	39.46	39.47	39.48	39.49	39.49	39.50	39.50	39.50
3	17.44	16.04	14.88	14.42	14.25	14.17	14.12	14.08	14.04	14.01	13.96	13.93	13.91	13.91	13.90
4	12.22	10.65	9.364	8.844	8.657	8.560	8.501	8.461	8.411	8.381	8.319	8.289	8.270	8.264	8.257
5	10.01	8.434	7.146	6.619	6.428	6.329	6.268	6.227	6.175	6.144	6.080	6.048	6.028	6.022	6.015
6	8.813	7.260	5.988	5.461	5.269	5.168	5.107	5.065	5.012	4.980	4.915	4.882	4.862	4.856	4.849
7	8.073	6.542	5.285	4.761	4.568	4.467	4.405	4.362	4.309	4.276	4.210	4.176	4.156	4.149	4.142
8	7.571	6.059	4.817	4.295	4.101	3.999	3.937	3.894	3.840	3.807	3.739	3.705	3.684	3.677	3.670
9	7.209	5.715	4.484	3.964	3.769	3.667	3.604	3.560	3.505	3.472	3.403	3.368	3.347	3.340	3.333
10	6.937	5.456	4.236	3.717	3.522	3.419	3.355	3.311	3.255	3.221	3.152	3.116	3.094	3.087	3.080
11	6.724	5.256	4.044	3.526	3.330	3.226	3.162	3.118	3.061	3.027	2.956	2.920	2.898	2.890	2.883
12	6.554	5.096	3.891	3.374	3.177	3.073	3.008	2.963	2.906	2.871	2.800	2.763	2.740	2.733	2.725
13	6.414	4.965	3.767	3.250	3.053	2.948	2.882	2.837	2.780	2.744	2.671	2.634	2.611	2.603	2.595
14	6.298	4.857	3.663	3.147	2.949	2.844	2.778	2.732	2.674	2.638	2.565	2.526	2.503	2.495	2.487
15	6.200	4.765	3.576	3.060	2.862	2.756	2.689	2.644	2.585	2.549	2.474	2.435	2.411	2.403	2.395
16	6.115	4.687	3.502	2.986	2.788	2.681	2.614	2.568	2.509	2.472	2.396	2.357	2.333	2.324	2.316
17	6.042	4.619	3.438	2.922	2.723	2.616	2.548	2.502	2.442	2.405	2.329	2.289	2.264	2.256	2.247
18	5.978	4.560	3.382	2.866	2.667	2.559	2.491	2.445	2.384	2.347	2.269	2.229	2.204	2.195	2.187
19	5.922	4.508	3.333	2.817	2.617	2.509	2.441	2.394	2.333	2.295	2.217	2.176	2.150	2.142	2.133
20	5.871	4.461	3.289	2.774	2.573	2.464	2.396	2.349	2.287	2.249	2.170	2.128	2.103	2.094	2.085
21	5.827	4.420	3.250	2.735	2.534	2.425	2.356	2.308	2.246	2.208	2.128	2.086	2.060	2.051	2.042
22	5.786	4.383	3.215	2.700	2.498	2.389	2.320	2.272	2.210	2.171	2.090	2.047	2.021	2.012	2.003
23	5.750	4.349	3.183	2.668	2.466	2.357	2.287	2.239	2.176	2.137	2.056	2.013	1.986	1.977	1.968
24	5.717	4.319	3.155	2.640	2.437	2.327	2.257	2.209	2.146	2.107	2.024	1.981	1.954	1.945	1.935
25	5.686	4.291	3.129	2.613	2.411	2.300	2.230	2.182	2.118	2.079	1.996	1.952	1.924	1.915	1.906
26	5.659	4.265	3.105	2.590	2.387	2.276	2.205	2.157	2.093	2.053	1.969	1.925	1.897	1.888	1.878
27	5.633	4.242	3.083	2.568	2.364	2.253	2.183	2.133	2.069	2.029	1.945	1.900	1.872	1.862	1.853
28	5.610	4.221	3.063	2.547	2.344	2.232	2.161	2.112	2.048	2.007	1.922	1.877	1.848	1.839	1.829
29	5.588	4.201	3.044	2.529	2.325	2.213	2.142	2.092	2.028	1.987	1.901	1.855	1.827	1.817	1.807
30	5.568	4.182	3.026	2.511	2.307	2.195	2.124	2.074	2.009	1.968	1.882	1.835	1.806	1.797	1.787
32	5.531	4.149	2.995	2.480	2.275	2.163	2.091	2.041	1.975	1.934	1.846	1.799	1.770	1.760	1.750
34	5.499	4.120	2.968	2.453	2.248	2.135	2.062	2.012	1.946	1.904	1.815	1.767	1.737	1.727	1.717
36	5.471	4.094	2.944	2.429	2.223	2.110	2.037	1.986	1.919	1.877	1.787	1.739	1.708	1.698	1.687
38	5.446	4.071	2.923	2.407	2.201	2.088	2.015	1.963	1.896	1.854	1.763	1.713	1.682	1.672	1.661
40	5.424	4.051	2.904	2.388	2.182	2.068	1.994	1.943	1.875	1.832	1.741	1.691	1.659	1.648	1.637
42	5.404	4.033	2.887	2.371	2.164	2.050	1.976	1.924	1.856	1.813	1.720	1.670	1.638	1.627	1.615
44	5.386	4.016	2.871	2.356	2.149	2.034	1.960	1.908	1.839	1.796	1.702	1.651	1.618	1.607	1.596
46	5.369	4.001	2.857	2.341	2.134	2.019	1.945	1.893	1.824	1.780	1.685	1.634	1.600	1.589	1.578
48	5.354	3.987	2.844	2.329	2.121	2.006	1.931	1.879	1.809	1.765	1.670	1.618	1.584	1.573	1.561
50	5.340	3.975	2.833	2.317	2.109	1.993	1.919	1.866	1.796	1.752	1.656	1.603	1.569	1.557	1.545
55	5.310	3.948	2.807	2.291	2.083	1.967	1.891	1.838	1.768	1.723	1.625	1.571	1.536	1.523	1.511
60	5.286	3.925	2.786	2.270	2.061	1.944	1.869	1.815	1.744	1.699	1.599	1.543	1.507	1.495	1.482
65	5.265	3.906	2.769	2.252	2.043	1.926	1.850	1.796	1.724	1.678	1.577	1.520	1.483	1.471	1.457
70	5.247	3.890	2.754	2.237	2.028	1.910	1.833	1.779	1.707	1.660	1.558	1.500	1.463	1.449	1.436
80	5.218	3.864	2.730	2.213	2.003	1.884	1.807	1.752	1.679	1.632	1.527	1.467	1.428	1.414	1.400
100	5.179	3.828	2.696	2.179	1.968	1.849	1.770	1.715	1.640	1.592	1.483	1.420	1.378	1.363	1.347
150	5.126	3.781	2.652	2.135	1.922	1.801	1.722	1.665	1.588	1.538	1.423	1.355	1.307	1.290	1.271
200	5.100	3.758	2.630	2.113	1.900	1.778	1.698	1.640	1.562	1.511	1.393	1.320	1.269	1.250	1.229
250	5.085	3.744	2.618	2.100	1.886	1.764	1.683	1.625	1.546	1.495	1.374	1.299	1.245	1.224	1.201
300	5.075	3.735	2.609	2.091	1.877	1.755	1.674	1.616	1.536	1.484	1.361	1.285	1.228	1.206	1.182
350	5.067	3.728	2.603	2.085	1.871	1.748	1.667	1.608	1.529	1.476	1.352	1.274	1.215	1.192	1.166
400	5.062	3.723	2.598	2.080	1.866	1.743	1.662	1.603	1.523	1.470	1.345	1.266	1.206	1.182	1.154
450	5.058	3.719	2.595	2.077	1.862	1.739	1.658	1.599	1.519	1.465	1.340	1.259	1.198	1.173	1.145
500	5.054	3.716	2.592	2.074	1.859	1.736	1.655	1.596	1.515	1.462	1.336	1.254	1.192	1.166	1.137
1000	5.039	3.703	2.579	2.061	1.846	1.722	1.640	1.581	1.499	1.445	1.316	1.230	1.162	1.132	1.094
∞	5.024	3.689	2.567	2.048	1.833	1.708	1.626	1.566	1.484	1.428	1.296	1.205	1.128	1.090	1.000

*DF = Denominator degrees of freedom

Table 13-11. *0.05 Critical Values for the Larger of Two Poisson Counts (M) Based on the Ratio Test, R = (M+1/2)/ (N+1/2), M > N*

Lower Count	M	Lower Count	M	Lower Count	M	Lower Count	M	Lower Count	M	Lower Count	M	Lower Count	M
1	7	51	73	101	131	151	188	201	243	251	297	301	352
2	9	52	74	102	132	152	189	202	244	252	298	302	353
3	10	53	76	103	134	153	190	203	245	253	300	303	354
4	12	54	77	104	135	154	191	204	246	254	301	304	355
5	14	55	78	105	136	155	192	205	247	255	302	305	356
6	15	56	79	106	137	156	193	206	248	256	303	306	357
7	17	57	80	107	138	157	194	207	249	257	304	307	358
8	18	58	82	108	139	158	195	208	250	258	305	308	359
9	20	59	83	109	140	159	196	209	252	259	306	309	360
10	21	60	84	110	142	160	198	210	253	260	307	310	361
11	23	61	85	111	143	161	199	211	254	261	308	311	362
12	24	62	86	112	144	162	200	212	255	262	309	312	363
13	26	63	87	113	145	163	201	213	256	263	310	313	364
14	27	64	89	114	146	164	202	214	257	264	311	314	366
15	28	65	90	115	147	165	203	215	258	265	313	315	367
16	30	66	91	116	148	166	204	216	259	266	314	316	368
17	31	67	92	117	149	167	205	217	260	267	315	317	369
18	32	68	93	118	151	168	206	218	261	268	316	318	370
19	34	69	94	119	152	169	207	219	262	269	317	319	371
20	35	70	96	120	153	170	209	220	264	270	318	320	372
21	36	71	97	121	154	171	210	221	265	271	319	321	373
22	37	72	98	122	155	172	211	222	266	272	320	322	374
23	39	73	99	123	156	173	212	223	267	273	321	323	375
24	40	74	100	124	157	174	213	224	268	274	322	324	376
25	41	75	101	125	158	175	214	225	269	275	323	325	377
26	43	76	103	126	160	176	215	226	270	276	324	326	378
27	44	77	104	127	161	177	216	227	271	277	326	327	380
28	45	78	105	128	162	178	217	228	272	278	327	328	381
29	46	79	106	129	163	179	219	229	273	279	328	329	382
30	48	80	107	130	164	180	220	230	274	280	329	330	383
31	49	81	108	131	165	181	221	231	276	281	330	331	384
32	50	82	110	132	166	182	222	232	277	282	331	332	385
33	51	83	111	133	167	183	223	233	278	283	332	333	386
34	53	84	112	134	169	184	224	234	279	284	333	334	387
35	54	85	113	135	170	185	225	235	280	285	334	335	388
36	55	86	114	136	171	186	226	236	281	286	335	336	389
37	56	87	115	137	172	187	227	237	282	287	336	337	390
38	58	88	116	138	173	188	228	238	283	288	337	338	391
39	59	89	118	139	174	189	230	239	284	289	339	339	392
40	60	90	119	140	175	190	231	240	285	290	340	340	394
41	61	91	120	141	176	191	232	241	286	291	341	341	395
42	62	92	121	142	177	192	233	242	288	292	342	342	396
43	64	93	122	143	179	193	234	243	289	293	343	343	397
44	65	94	123	144	180	194	235	244	290	294	344	344	398
45	66	95	124	145	181	195	236	245	291	295	345	345	399
46	67	96	126	146	182	196	237	246	292	296	346	346	400
47	68	97	127	147	183	197	238	247	293	297	347	347	401
48	70	98	128	148	184	198	239	248	294	298	348	348	402
49	71	99	129	149	185	199	241	249	295	299	349	349	403
50	72	100	130	150	186	200	242	250	296	300	350	350	404

Table 13-12. *Lower and Upper 95% Confidence Limits for the Mean of a Poisson Variate*

Count	Lower Limit	Upper Limit	Count	Lower Limit	Upper Limit	Count	Lower Limit	Upper Limit	Count	Lower Limit	Upper Limit	Count	Lower Limit	Upper Limit	Count	Lower Limit	Upper Limit
0	0	3.689	50	37.11	65.92	100	81.36	121.6	150	127.0	176.0	200	173.2	229.7	250	220.0	283.0
1	.0253	5.572	51	37.97	67.06	101	82.27	122.7	151	127.9	177.1	201	174.2	230.8	251	220.9	284.0
2	.2422	7.225	52	38.84	68.19	102	83.17	123.8	152	128.8	178.2	202	175.1	231.9	252	221.8	285.1
3	.6187	8.767	53	39.70	69.33	103	84.07	124.9	153	129.7	179.3	203	176.0	232.9	253	222.8	286.2
4	1.090	10.24	54	40.57	70.46	104	84.98	126.0	154	130.6	180.3	204	177.0	234.0	254	223.7	287.2
5	1.623	11.67	55	41.43	71.59	105	85.88	127.1	155	131.6	181.4	205	177.9	235.1	255	224.7	288.3
6	2.202	13.06	56	42.30	72.72	106	86.78	128.2	156	132.5	182.5	206	178.8	236.1	256	225.6	289.4
7	2.814	14.42	57	43.17	73.85	107	87.69	129.3	157	133.4	183.6	207	179.8	237.2	257	226.5	290.4
8	3.454	15.76	58	44.04	74.98	108	88.59	130.4	158	134.3	184.6	208	180.7	238.3	258	227.5	291.5
9	4.115	17.08	59	44.91	76.11	109	89.50	131.5	159	135.2	185.7	209	181.6	239.3	259	228.4	292.5
10	4.795	18.39	60	45.79	77.23	110	90.41	132.6	160	136.2	186.8	210	182.6	240.4	260	229.4	293.6
11	5.491	19.68	61	46.66	78.36	111	91.31	133.7	161	137.1	187.9	211	183.5	241.5	261	230.3	294.7
12	6.201	20.96	62	47.54	79.48	112	92.22	134.8	162	138.0	189.0	212	184.4	242.5	262	231.2	295.7
13	6.922	22.23	63	48.41	80.60	113	93.13	135.9	163	138.9	190.0	213	185.4	243.6	263	232.2	296.8
14	7.654	23.49	64	49.29	81.73	114	94.04	136.9	164	139.9	191.1	214	186.3	244.7	264	233.1	297.8
15	8.395	24.74	65	50.17	82.85	115	94.94	138.0	165	140.8	192.2	215	187.2	245.7	265	234.1	298.9
16	9.145	25.98	66	51.04	83.97	116	95.85	139.1	166	141.7	193.3	216	188.2	246.8	266	235.0	300.0
17	9.903	27.22	67	51.92	85.09	117	96.76	140.2	167	142.6	194.3	217	189.1	247.9	267	235.9	301.0
18	10.67	28.45	68	52.80	86.21	118	97.67	141.3	168	143.6	195.4	218	190.0	248.9	268	236.9	302.1
19	11.44	29.67	69	53.69	87.32	119	98.58	142.4	169	144.5	196.5	219	191.0	250.0	269	237.8	303.1
20	12.22	30.89	70	54.57	88.44	120	99.49	143.5	170	145.4	197.6	220	191.9	251.1	270	238.8	304.2
21	13.00	32.10	71	55.45	89.56	121	100.4	144.6	171	146.3	198.6	221	192.8	252.1	271	239.7	305.3
22	13.79	33.31	72	56.34	90.67	122	101.3	145.7	172	147.3	199.7	222	193.8	253.2	272	240.6	306.3
23	14.58	34.51	73	57.22	91.79	123	102.2	146.8	173	148.2	200.8	223	194.7	254.3	273	241.6	307.4
24	15.38	35.71	74	58.11	92.90	124	103.1	147.8	174	149.1	201.9	224	195.6	255.3	274	242.5	308.4
25	16.18	36.90	75	58.99	94.01	125	104.0	148.9	175	150.0	202.9	225	196.6	256.4	275	243.5	309.5
26	16.98	38.10	76	59.88	95.13	126	105.0	150.0	176	151.0	204.0	226	197.5	257.5	276	244.4	310.6
27	17.79	39.28	77	60.77	96.24	127	105.9	151.1	177	151.9	205.1	227	198.4	258.5	277	245.3	311.6
28	18.61	40.47	78	61.66	97.35	128	106.8	152.2	178	152.8	206.2	228	199.4	259.6	278	246.3	312.7
29	19.42	41.65	79	62.55	98.46	129	107.7	153.3	179	153.7	207.2	229	200.3	260.7	279	247.2	313.7
30	20.24	42.83	80	63.44	99.57	130	108.6	154.4	180	154.7	208.3	230	201.2	261.7	280	248.2	314.8
31	21.06	44.00	81	64.33	100.7	131	109.5	155.4	181	155.6	209.4	231	202.2	262.8	281	249.1	315.9
32	21.89	45.17	82	65.22	101.8	132	110.4	156.5	182	156.5	210.4	232	203.1	263.9	282	250.0	316.9
33	22.72	46.34	83	66.11	102.9	133	111.4	157.6	183	157.4	211.5	233	204.0	264.9	283	251.0	318.0
34	23.55	47.51	84	67.00	104.0	134	112.3	158.7	184	158.4	212.6	234	205.0	266.0	284	251.9	319.0
35	24.38	48.68	85	67.89	105.1	135	113.2	159.8	185	159.3	213.7	235	205.9	267.0	285	252.9	320.1
36	25.21	49.84	86	68.79	106.2	136	114.1	160.9	186	160.2	214.7	236	206.8	268.1	286	253.8	321.1
37	26.05	51.00	87	69.68	107.3	137	115.0	162.0	187	161.2	215.8	237	207.8	269.2	287	254.8	322.2
38	26.89	52.16	88	70.58	108.4	138	115.9	163.0	188	162.1	216.9	238	208.7	270.2	288	255.7	323.3
39	27.73	53.31	89	71.47	109.5	139	116.9	164.1	189	163.0	218.0	239	209.7	271.3	289	256.6	324.3
40	28.58	54.47	90	72.37	110.6	140	117.8	165.2	190	163.9	219.0	240	210.6	272.4	290	257.6	325.4
41	29.42	55.62	91	73.27	111.7	141	118.7	166.3	191	164.9	220.1	241	211.5	273.4	291	258.5	326.4
42	30.27	56.77	92	74.16	112.8	142	119.6	167.4	192	165.8	221.2	242	212.5	274.5	292	259.5	327.5
43	31.12	57.92	93	75.06	113.9	143	120.5	168.5	193	166.7	222.2	243	213.4	275.6	293	260.4	328.5
44	31.97	59.07	94	75.96	115.0	144	121.4	169.5	194	167.7	223.3	244	214.3	276.6	294	261.4	329.6
45	32.82	60.21	95	76.86	116.1	145	122.4	170.6	195	168.6	224.4	245	215.3	277.7	295	262.3	330.7
46	33.68	61.36	96	77.76	117.2	146	123.3	171.7	196	169.5	225.4	246	216.2	278.7	296	263.2	331.7
47	34.53	62.50	97	78.66	118.3	147	124.2	172.8	197	170.4	226.5	247	217.2	279.8	297	264.2	332.8
48	35.39	63.64	98	79.56	119.4	148	125.1	173.9	198	171.4	227.6	248	218.1	280.9	298	265.1	333.8
49	36.25	64.78	99	80.46	120.5	149	126.0	174.9	199	172.3	228.7	249	219.0	281.9	299	266.1	334.9

Table 13-13. *Lower and Upper Simultaneous 95% Confidence Limits for the Means of Several Poisson Variates*

 # Groups = 2

Count	Lower Limit	Upper Limit	Count	Lower Limit	Upper Limit	Count	Lower Limit	Upper Limit	Count	Lower Limit	Upper Limit	Count	Lower Limit	Upper Limit	Count	Lower Limit	Upper Limit
0	0	4.382	50	35.51	68.33	100	78.94	124.9	150	123.9	179.9	200	169.7	234.1	250	215.9	287.8
1	.0126	6.381	51	36.35	69.49	101	79.83	126.0	151	124.8	181.0	201	170.6	235.2	251	216.8	288.9
2	.1671	8.122	52	37.19	70.64	102	80.72	127.1	152	125.7	182.1	202	171.5	236.3	252	217.8	290.0
3	.4741	9.739	53	38.04	71.79	103	81.60	128.2	153	126.6	183.1	203	172.4	237.3	253	218.7	291.1
4	.8801	11.28	54	38.88	72.95	104	82.49	129.3	154	127.5	184.2	204	173.3	238.4	254	219.6	292.1
5	1.354	12.77	55	39.73	74.10	105	83.38	130.4	155	128.4	185.3	205	174.3	239.5	255	220.6	293.2
6	1.876	14.21	56	40.58	75.25	106	84.28	131.5	156	129.4	186.4	206	175.2	240.6	256	221.5	294.3
7	2.437	15.63	57	41.43	76.39	107	85.17	132.6	157	130.3	187.5	207	176.1	241.7	257	222.4	295.3
8	3.027	17.01	58	42.29	77.54	108	86.06	133.7	158	131.2	188.6	208	177.0	242.7	258	223.3	296.4
9	3.642	18.38	59	43.14	78.69	109	86.95	134.8	159	132.1	189.7	209	177.9	243.8	259	224.3	297.5
10	4.278	19.73	60	43.99	79.83	110	87.84	135.9	160	133.0	190.8	210	178.9	244.9	260	225.2	298.5
11	4.932	21.06	61	44.85	80.97	111	88.74	137.0	161	133.9	191.9	211	179.8	246.0	261	226.1	299.6
12	5.601	22.38	62	45.71	82.11	112	89.63	138.2	162	134.8	192.9	212	180.7	247.0	262	227.1	300.7
13	6.283	23.69	63	46.56	83.26	113	90.53	139.3	163	135.7	194.0	213	181.6	248.1	263	228.0	301.8
14	6.978	24.98	64	47.42	84.40	114	91.42	140.4	164	136.6	195.1	214	182.6	249.2	264	228.9	302.8
15	7.683	26.27	65	48.28	85.53	115	92.32	141.5	165	137.6	196.2	215	183.5	250.3	265	229.9	303.9
16	8.398	27.55	66	49.15	86.67	116	93.21	142.6	166	138.5	197.3	216	184.4	251.4	266	230.8	305.0
17	9.122	28.82	67	50.01	87.81	117	94.11	143.7	167	139.4	198.4	217	185.3	252.4	267	231.7	306.0
18	9.853	30.08	68	50.87	88.94	118	95.00	144.8	168	140.3	199.5	218	186.3	253.5	268	232.7	307.1
19	10.59	31.33	69	51.74	90.08	119	95.90	145.9	169	141.2	200.6	219	187.2	254.6	269	233.6	308.2
20	11.34	32.58	70	52.60	91.21	120	96.80	147.0	170	142.1	201.6	220	188.1	255.7	270	234.5	309.2
21	12.09	33.82	71	53.47	92.34	121	97.70	148.1	171	143.0	202.7	221	189.0	256.7	271	235.5	310.3
22	12.85	35.06	72	54.34	93.48	122	98.59	149.2	172	144.0	203.8	222	190.0	257.8	272	236.4	311.4
23	13.61	36.29	73	55.20	94.61	123	99.49	150.3	173	144.9	204.9	223	190.9	258.9	273	237.3	312.4
24	14.38	37.52	74	56.07	95.74	124	100.4	151.4	174	145.8	206.0	224	191.8	260.0	274	238.2	313.5
25	15.15	38.74	75	56.94	96.87	125	101.3	152.5	175	146.7	207.1	225	192.7	261.0	275	239.2	314.6
26	15.93	39.96	76	57.81	97.99	126	102.2	153.6	176	147.6	208.2	226	193.7	262.1	276	240.1	315.6
27	16.71	41.17	77	58.69	99.12	127	103.1	154.7	177	148.5	209.2	227	194.6	263.2	277	241.0	316.7
28	17.50	42.39	78	59.56	100.2	128	104.0	155.8	178	149.4	210.3	228	195.5	264.3	278	242.0	317.8
29	18.29	43.59	79	60.43	101.4	129	104.9	156.9	179	150.4	211.4	229	196.4	265.3	279	242.9	318.8
30	19.08	44.80	80	61.31	102.5	130	105.8	158.0	180	151.3	212.5	230	197.4	266.4	280	243.8	319.9
31	19.88	46.00	81	62.18	103.6	131	106.7	159.1	181	152.2	213.6	231	198.3	267.5	281	244.8	321.0
32	20.68	47.19	82	63.06	104.7	132	107.6	160.2	182	153.1	214.7	232	199.2	268.5	282	245.7	322.0
33	21.48	48.39	83	63.93	105.9	133	108.5	161.3	183	154.0	215.7	233	200.1	269.6	283	246.6	323.1
34	22.29	49.58	84	64.81	107.0	134	109.4	162.4	184	154.9	216.8	234	201.1	270.7	284	247.6	324.2
35	23.10	50.77	85	65.69	108.1	135	110.3	163.5	185	155.9	217.9	235	202.0	271.8	285	248.5	325.2
36	23.91	51.95	86	66.57	109.2	136	111.2	164.6	186	156.8	219.0	236	202.9	272.8	286	249.4	326.3
37	24.72	53.14	87	67.45	110.4	137	112.1	165.7	187	157.7	220.1	237	203.8	273.9	287	250.4	327.4
38	25.54	54.32	88	68.33	111.5	138	113.0	166.8	188	158.6	221.1	238	204.8	275.0	288	251.3	328.4
39	26.36	55.50	89	69.21	112.6	139	113.9	167.9	189	159.5	222.2	239	205.7	276.1	289	252.2	329.5
40	27.18	56.67	90	70.09	113.7	140	114.8	168.9	190	160.5	223.3	240	206.6	277.1	290	253.2	330.6
41	28.01	57.85	91	70.97	114.8	141	115.7	170.0	191	161.4	224.4	241	207.6	278.2	291	254.1	331.6
42	28.83	59.02	92	71.85	115.9	142	116.6	171.1	192	162.3	225.5	242	208.5	279.3	292	255.0	332.7
43	29.66	60.19	93	72.74	117.1	143	117.5	172.2	193	163.2	226.6	243	209.4	280.3	293	256.0	333.8
44	30.49	61.36	94	73.62	118.2	144	118.5	173.3	194	164.1	227.6	244	210.3	281.4	294	256.9	334.8
45	31.32	62.52	95	74.51	119.3	145	119.4	174.4	195	165.0	228.7	245	211.3	282.5	295	257.9	335.9
46	32.15	63.69	96	75.39	120.4	146	120.3	175.5	196	166.0	229.8	246	212.2	283.6	296	258.8	337.0
47	32.99	64.85	97	76.28	121.5	147	121.2	176.6	197	166.9	230.9	247	213.1	284.6	297	259.7	338.0
48	33.83	66.01	98	77.16	122.6	148	122.1	177.7	198	167.8	232.0	248	214.1	285.7	298	260.7	339.1
49	34.67	67.17	99	78.05	123.7	149	123.0	178.8	199	168.7	233.0	249	215.0	286.8	299	261.6	340.2

(continued)

Table 13-13. *(continued)*

 # Groups = 3

Count	Lower Limit	Upper Limit	Count	Lower Limit	Upper Limit	Count	Lower Limit	Upper Limit	Count	Lower Limit	Upper Limit	Count	Lower Limit	Upper Limit	Count	Lower Limit	Upper Limit
0	0	4.787	50	34.66	69.66	100	77.64	126.6	150	122.3	182.0	200	167.7	236.5	250	213.7	290.5
1	.0084	6.848	51	35.49	70.83	101	78.52	127.7	151	123.2	183.1	201	168.6	237.6	251	214.7	291.6
2	.1350	8.636	52	36.32	71.99	102	79.41	128.9	152	124.1	184.2	202	169.6	238.7	252	215.6	292.6
3	.4075	10.29	53	37.16	73.16	103	80.29	130.0	153	125.0	185.3	203	170.5	239.8	253	216.5	293.7
4	.7800	11.87	54	37.99	74.32	104	81.17	131.1	154	125.9	186.4	204	171.4	240.8	254	217.4	294.8
5	1.222	13.39	55	38.83	75.48	105	82.05	132.2	155	126.8	187.5	205	172.3	241.9	255	218.4	295.9
6	1.715	14.86	56	39.67	76.64	106	82.94	133.3	156	127.7	188.6	206	173.2	243.0	256	219.3	296.9
7	2.248	16.30	57	40.51	77.80	107	83.82	134.4	157	128.6	189.7	207	174.1	244.1	257	220.2	298.0
8	2.812	17.72	58	41.35	78.95	108	84.70	135.6	158	129.5	190.8	208	175.1	245.2	258	221.1	299.1
9	3.403	19.11	59	42.20	80.11	109	85.59	136.7	159	130.4	191.9	209	176.0	246.3	259	222.1	300.2
10	4.015	20.48	60	43.04	81.26	110	86.48	137.8	160	131.3	192.9	210	176.9	247.3	260	223.0	301.2
11	4.646	21.83	61	43.89	82.41	111	87.36	138.9	161	132.2	194.0	211	177.8	248.4	261	223.9	302.3
12	5.293	23.18	62	44.73	83.57	112	88.25	140.0	162	133.1	195.1	212	178.7	249.5	262	224.8	303.4
13	5.955	24.50	63	45.58	84.72	113	89.14	141.1	163	134.0	196.2	213	179.6	250.6	263	225.8	304.5
14	6.630	25.82	64	46.43	85.87	114	90.02	142.2	164	134.9	197.3	214	180.6	251.7	264	226.7	305.5
15	7.315	27.12	65	47.28	87.01	115	90.91	143.4	165	135.8	198.4	215	181.5	252.8	265	227.6	306.6
16	8.011	28.42	66	48.14	88.16	116	91.80	144.5	166	136.7	199.5	216	182.4	253.8	266	228.5	307.7
17	8.717	29.71	67	48.99	89.31	117	92.69	145.6	167	137.6	200.6	217	183.3	254.9	267	229.5	308.8
18	9.430	30.99	68	49.84	90.45	118	93.58	146.7	168	138.6	201.7	218	184.2	256.0	268	230.4	309.8
19	10.15	32.26	69	50.70	91.59	119	94.47	147.8	169	139.5	202.8	219	185.2	257.1	269	231.3	310.9
20	10.88	33.52	70	51.56	92.74	120	95.36	148.9	170	140.4	203.9	220	186.1	258.2	270	232.2	312.0
21	11.62	34.78	71	52.41	93.88	121	96.25	150.0	171	141.3	205.0	221	187.0	259.2	271	233.2	313.1
22	12.36	36.03	72	53.27	95.02	122	97.14	151.1	172	142.2	206.1	222	187.9	260.3	272	234.1	314.1
23	13.11	37.28	73	54.13	96.16	123	98.03	152.2	173	143.1	207.1	223	188.8	261.4	273	235.0	315.2
24	13.86	38.52	74	54.99	97.30	124	98.93	153.3	174	144.0	208.2	224	189.8	262.5	274	236.0	316.3
25	14.62	39.76	75	55.85	98.44	125	99.82	154.4	175	144.9	209.3	225	190.7	263.6	275	236.9	317.3
26	15.38	41.00	76	56.71	99.57	126	100.7	155.5	176	145.8	210.4	226	191.6	264.6	276	237.8	318.4
27	16.15	42.22	77	57.58	100.7	127	101.6	156.7	177	146.7	211.5	227	192.5	265.7	277	238.7	319.5
28	16.92	43.45	78	58.44	101.8	128	102.5	157.8	178	147.6	212.6	228	193.4	266.8	278	239.7	320.6
29	17.69	44.67	79	59.31	103.0	129	103.4	158.9	179	148.6	213.7	229	194.4	267.9	279	240.6	321.6
30	18.47	45.89	80	60.17	104.1	130	104.3	160.0	180	149.5	214.8	230	195.3	269.0	280	241.5	322.7
31	19.26	47.10	81	61.04	105.2	131	105.2	161.1	181	150.4	215.9	231	196.2	270.0	281	242.5	323.8
32	20.04	48.31	82	61.91	106.4	132	106.1	162.2	182	151.3	217.0	232	197.1	271.1	282	243.4	324.8
33	20.83	49.52	83	62.77	107.5	133	107.0	163.3	183	152.2	218.0	233	198.0	272.2	283	244.3	325.9
34	21.63	50.72	84	63.64	108.6	134	107.9	164.4	184	153.1	219.1	234	199.0	273.3	284	245.2	327.0
35	22.42	51.92	85	64.51	109.8	135	108.8	165.5	185	154.0	220.2	235	199.9	274.3	285	246.2	328.1
36	23.22	53.12	86	65.38	110.9	136	109.7	166.6	186	154.9	221.3	236	200.8	275.4	286	247.1	329.1
37	24.02	54.32	87	66.25	112.0	137	110.6	167.7	187	155.8	222.4	237	201.7	276.5	287	248.0	330.2
38	24.83	55.51	88	67.13	113.2	138	111.5	168.8	188	156.8	223.5	238	202.6	277.6	288	249.0	331.3
39	25.64	56.70	89	68.00	114.3	139	112.4	169.9	189	157.7	224.6	239	203.6	278.7	289	249.9	332.3
40	26.45	57.89	90	68.87	115.4	140	113.3	171.0	190	158.6	225.7	240	204.5	279.7	290	250.8	333.4
41	27.26	59.08	91	69.75	116.5	141	114.2	172.1	191	159.5	226.7	241	205.4	280.8	291	251.7	334.5
42	28.07	60.26	92	70.62	117.7	142	115.1	173.2	192	160.4	227.8	242	206.3	281.9	292	252.7	335.5
43	28.89	61.44	93	71.50	118.8	143	116.0	174.3	193	161.3	228.9	243	207.3	283.0	293	253.6	336.6
44	29.71	62.62	94	72.37	119.9	144	116.9	175.4	194	162.2	230.0	244	208.2	284.0	294	254.5	337.7
45	30.53	63.80	95	73.25	121.0	145	117.8	176.5	195	163.2	231.1	245	209.1	285.1	295	255.5	338.8
46	31.35	64.97	96	74.13	122.1	146	118.7	177.6	196	164.1	232.2	246	210.0	286.2	296	256.4	339.8
47	32.17	66.15	97	75.01	123.3	147	119.6	178.7	197	165.0	233.3	247	211.0	287.3	297	257.3	340.9
48	33.00	67.32	98	75.88	124.4	148	120.5	179.8	198	165.9	234.3	248	211.9	288.3	298	258.3	342.0
49	33.83	68.49	99	76.76	125.5	149	121.4	180.9	199	166.8	235.4	249	212.8	289.4	299	259.2	343.0

(continued)

Table 13-13. (continued)

 # Groups = 4

Count	Lower Limit	Upper Limit	Count	Lower Limit	Upper Limit	Count	Lower Limit	Upper Limit	Count	Lower Limit	Upper Limit	Count	Lower Limit	Upper Limit	Count	Lower Limit	Upper Limit
0	0	5.075	50	34.09	70.57	100	76.77	127.8	150	121.2	183.4	200	166.4	238.2	250	212.3	292.3
1	.0063	7.176	51	34.91	71.75	101	77.65	129.0	151	122.1	184.5	201	167.3	239.2	251	213.2	293.4
2	.1162	8.996	52	35.74	72.92	102	78.52	130.1	152	123.0	185.6	202	168.2	240.3	252	214.1	294.5
3	.3665	10.68	53	36.57	74.09	103	79.40	131.2	153	123.9	186.7	203	169.2	241.4	253	215.0	295.5
4	.7170	12.28	54	37.40	75.26	104	80.28	132.3	154	124.8	187.8	204	170.1	242.5	254	215.9	296.6
5	1.138	13.82	55	38.23	76.43	105	81.16	133.5	155	125.7	188.9	205	171.0	243.6	255	216.9	297.7
6	1.612	15.31	56	39.06	77.59	106	82.03	134.6	156	126.6	190.0	206	171.9	244.7	256	217.8	298.8
7	2.126	16.77	57	39.89	78.76	107	82.91	135.7	157	127.5	191.1	207	172.8	245.8	257	218.7	299.9
8	2.673	18.20	58	40.73	79.92	108	83.79	136.8	158	128.4	192.2	208	173.7	246.9	258	219.6	300.9
9	3.247	19.61	59	41.56	81.08	109	84.67	137.9	159	129.3	193.3	209	174.6	247.9	259	220.6	302.0
10	3.843	21.00	60	42.40	82.24	110	85.55	139.1	160	130.2	194.4	210	175.6	249.0	260	221.5	303.1
11	4.459	22.37	61	43.24	83.40	111	86.43	140.2	161	131.1	195.5	211	176.5	250.1	261	222.4	304.2
12	5.092	23.73	62	44.08	84.56	112	87.32	141.3	162	132.0	196.6	212	177.4	251.2	262	223.3	305.2
13	5.739	25.07	63	44.92	85.72	113	88.20	142.4	163	132.9	197.7	213	178.3	252.3	263	224.2	306.3
14	6.400	26.40	64	45.77	86.87	114	89.08	143.5	164	133.8	198.8	214	179.2	253.4	264	225.2	307.4
15	7.072	27.71	65	46.61	88.03	115	89.96	144.6	165	134.7	199.9	215	180.1	254.5	265	226.1	308.5
16	7.756	29.02	66	47.46	89.18	116	90.85	145.8	166	135.6	201.0	216	181.0	255.5	266	227.0	309.6
17	8.449	30.32	67	48.31	90.33	117	91.73	146.9	167	136.5	202.1	217	182.0	256.6	267	227.9	310.6
18	9.150	31.61	68	49.15	91.48	118	92.62	148.0	168	137.4	203.2	218	182.9	257.7	268	228.9	311.7
19	9.860	32.90	69	50.00	92.63	119	93.50	149.1	169	138.3	204.3	219	183.8	258.8	269	229.8	312.8
20	10.58	34.17	70	50.85	93.78	120	94.39	150.2	170	139.2	205.4	220	184.7	259.9	270	230.7	313.9
21	11.30	35.44	71	51.70	94.93	121	95.27	151.3	171	140.1	206.5	221	185.6	261.0	271	231.6	314.9
22	12.03	36.71	72	52.56	96.08	122	96.16	152.4	172	141.0	207.6	222	186.5	262.0	272	232.6	316.0
23	12.77	37.96	73	53.41	97.22	123	97.05	153.6	173	141.9	208.7	223	187.5	263.1	273	233.5	317.1
24	13.51	39.22	74	54.26	98.37	124	97.94	154.7	174	142.8	209.8	224	188.4	264.2	274	234.4	318.2
25	14.26	40.47	75	55.12	99.51	125	98.82	155.8	175	143.7	210.9	225	189.3	265.3	275	235.3	319.2
26	15.01	41.71	76	55.98	100.7	126	99.71	156.9	176	144.6	212.0	226	190.2	266.4	276	236.3	320.3
27	15.77	42.95	77	56.83	101.8	127	100.6	158.0	177	145.5	213.1	227	191.1	267.5	277	237.2	321.4
28	16.53	44.18	78	57.69	102.9	128	101.5	159.1	178	146.4	214.2	228	192.0	268.5	278	238.1	322.5
29	17.30	45.41	79	58.55	104.1	129	102.4	160.2	179	147.3	215.3	229	193.0	269.6	279	239.0	323.5
30	18.07	46.64	80	59.41	105.2	130	103.3	161.3	180	148.2	216.3	230	193.9	270.7	280	240.0	324.6
31	18.84	47.86	81	60.27	106.4	131	104.2	162.4	181	149.1	217.4	231	194.8	271.8	281	240.9	325.7
32	19.62	49.08	82	61.13	107.5	132	105.1	163.5	182	150.1	218.5	232	195.7	272.9	282	241.8	326.8
33	20.40	50.30	83	61.99	108.6	133	105.9	164.7	183	151.0	219.6	233	196.6	273.9	283	242.7	327.8
34	21.19	51.51	84	62.86	109.8	134	106.8	165.8	184	151.9	220.7	234	197.5	275.0	284	243.7	328.9
35	21.97	52.72	85	63.72	110.9	135	107.7	166.9	185	152.8	221.8	235	198.5	276.1	285	244.6	330.0
36	22.76	53.93	86	64.59	112.0	136	108.6	168.0	186	153.7	222.9	236	199.4	277.2	286	245.5	331.1
37	23.56	55.13	87	65.45	113.2	137	109.5	169.1	187	154.6	224.0	237	200.3	278.3	287	246.4	332.1
38	24.35	56.33	88	66.32	114.3	138	110.4	170.2	188	155.5	225.1	238	201.2	279.4	288	247.4	333.2
39	25.15	57.53	89	67.19	115.4	139	111.3	171.3	189	156.4	226.2	239	202.1	280.4	289	248.3	334.3
40	25.95	58.73	90	68.05	116.6	140	112.2	172.4	190	157.3	227.3	240	203.1	281.5	290	249.2	335.4
41	26.76	59.92	91	68.92	117.7	141	113.1	173.5	191	158.2	228.4	241	204.0	282.6	291	250.1	336.4
42	27.56	61.11	92	69.79	118.8	142	114.0	174.6	192	159.1	229.4	242	204.9	283.7	292	251.1	337.5
43	28.37	62.30	93	70.66	120.0	143	114.9	175.7	193	160.0	230.5	243	205.8	284.8	293	252.0	338.6
44	29.18	63.49	94	71.53	121.1	144	115.8	176.8	194	161.0	231.6	244	206.7	285.8	294	252.9	339.6
45	29.99	64.68	95	72.40	122.2	145	116.7	177.9	195	161.9	232.7	245	207.7	286.9	295	253.8	340.7
46	30.81	65.86	96	73.28	123.3	146	117.6	179.0	196	162.8	233.8	246	208.6	288.0	296	254.8	341.8
47	31.63	67.04	97	74.15	124.5	147	118.5	180.1	197	163.7	234.9	247	209.5	289.1	297	255.7	342.9
48	32.45	68.22	98	75.02	125.6	148	119.4	181.2	198	164.6	236.0	248	210.4	290.2	298	256.6	343.9
49	33.27	69.40	99	75.90	126.7	149	120.3	182.3	199	165.5	237.1	249	211.3	291.2	299	257.6	345.0

(continued)

Table 13-13. *(continued)*

 # Groups = 5

Count	Lower Limit	Upper Limit	Count	Lower Limit	Upper Limit	Count	Lower Limit	Upper Limit	Count	Lower Limit	Upper Limit	Count	Lower Limit	Upper Limit	Count	Lower Limit	Upper Limit
0	0	5.298	50	33.66	71.27	100	76.12	128.8	150	120.3	184.5	200	165.5	239.4	250	211.2	293.7
1	.0050	7.430	51	34.48	72.45	101	76.99	129.9	151	121.2	185.6	201	166.4	240.5	251	212.1	294.8
2	.1035	9.274	52	35.30	73.62	102	77.86	131.0	152	122.1	186.7	202	167.3	241.6	252	213.0	295.8
3	.3379	10.98	53	36.13	74.80	103	78.74	132.1	153	123.0	187.8	203	168.2	242.7	253	213.9	296.9
4	.6722	12.59	54	36.95	75.97	104	79.61	133.3	154	123.9	188.9	204	169.1	243.8	254	214.8	298.0
5	1.078	14.15	55	37.78	77.15	105	80.48	134.4	155	124.8	190.0	205	170.0	244.8	255	215.7	299.1
6	1.537	15.66	56	38.60	78.32	106	81.36	135.5	156	125.7	191.2	206	170.9	245.9	256	216.7	300.2
7	2.037	17.13	57	39.43	79.49	107	82.23	136.6	157	126.6	192.3	207	171.8	247.0	257	217.6	301.2
8	2.571	18.58	58	40.26	80.66	108	83.11	137.8	158	127.5	193.4	208	172.7	248.1	258	218.5	302.3
9	3.132	20.00	59	41.09	81.82	109	83.99	138.9	159	128.4	194.5	209	173.6	249.2	259	219.4	303.4
10	3.717	21.40	60	41.93	82.99	110	84.86	140.0	160	129.3	195.6	210	174.6	250.3	260	220.3	304.5
11	4.321	22.78	61	42.76	84.15	111	85.74	141.1	161	130.2	196.7	211	175.5	251.4	261	221.3	305.6
12	4.943	24.14	62	43.60	85.32	112	86.62	142.3	162	131.1	197.8	212	176.4	252.5	262	222.2	306.6
13	5.580	25.50	63	44.43	86.48	113	87.50	143.4	163	132.0	198.9	213	177.3	253.6	263	223.1	307.7
14	6.231	26.84	64	45.27	87.64	114	88.38	144.5	164	132.9	200.0	214	178.2	254.6	264	224.0	308.8
15	6.893	28.16	65	46.11	88.80	115	89.26	145.6	165	133.8	201.1	215	179.1	255.7	265	224.9	309.9
16	7.567	29.48	66	46.95	89.96	116	90.14	146.7	166	134.7	202.2	216	180.0	256.8	266	225.9	311.0
17	8.251	30.79	67	47.79	91.11	117	91.02	147.9	167	135.6	203.3	217	180.9	257.9	267	226.8	312.0
18	8.943	32.09	68	48.64	92.27	118	91.90	149.0	168	136.5	204.4	218	181.8	259.0	268	227.7	313.1
19	9.644	33.38	69	49.48	93.42	119	92.78	150.1	169	137.4	205.5	219	182.8	260.1	269	228.6	314.2
20	10.35	34.67	70	50.33	94.58	120	93.66	151.2	170	138.3	206.6	220	183.7	261.2	270	229.6	315.3
21	11.07	35.95	71	51.17	95.73	121	94.54	152.3	171	139.2	207.7	221	184.6	262.3	271	230.5	316.4
22	11.79	37.22	72	52.02	96.88	122	95.43	153.4	172	140.1	208.8	222	185.5	263.3	272	231.4	317.4
23	12.52	38.48	73	52.87	98.03	123	96.31	154.6	173	141.0	209.9	223	186.4	264.4	273	232.3	318.5
24	13.26	39.74	74	53.72	99.18	124	97.20	155.7	174	141.9	211.0	224	187.3	265.5	274	233.2	319.6
25	14.00	41.00	75	54.57	100.3	125	98.08	156.8	175	142.8	212.0	225	188.2	266.6	275	234.2	320.7
26	14.74	42.25	76	55.42	101.5	126	98.97	157.9	176	143.7	213.1	226	189.2	267.7	276	235.1	321.7
27	15.49	43.50	77	56.28	102.6	127	99.85	159.0	177	144.6	214.2	227	190.1	268.8	277	236.0	322.8
28	16.25	44.74	78	57.13	103.8	128	100.7	160.1	178	145.5	215.3	228	191.0	269.9	278	236.9	323.9
29	17.00	45.98	79	57.98	104.9	129	101.6	161.2	179	146.4	216.4	229	191.9	270.9	279	237.9	325.0
30	17.77	47.21	80	58.84	106.1	130	102.5	162.4	180	147.3	217.5	230	192.8	272.0	280	238.8	326.1
31	18.53	48.44	81	59.70	107.2	131	103.4	163.5	181	148.2	218.6	231	193.7	273.1	281	239.7	327.1
32	19.30	49.67	82	60.55	108.3	132	104.3	164.6	182	149.1	219.7	232	194.6	274.2	282	240.6	328.2
33	20.08	50.89	83	61.41	109.5	133	105.2	165.7	183	150.0	220.8	233	195.6	275.3	283	241.5	329.3
34	20.86	52.11	84	62.27	110.6	134	106.1	166.8	184	150.9	221.9	234	196.5	276.4	284	242.5	330.4
35	21.64	53.32	85	63.13	111.8	135	107.0	167.9	185	151.8	223.0	235	197.4	277.4	285	243.4	331.4
36	22.42	54.54	86	63.99	112.9	136	107.8	169.0	186	152.7	224.1	236	198.3	278.5	286	244.3	332.5
37	23.21	55.75	87	64.85	114.0	137	108.7	170.1	187	153.7	225.2	237	199.2	279.6	287	245.2	333.6
38	24.00	56.96	88	65.72	115.2	138	109.6	171.2	188	154.6	226.3	238	200.1	280.7	288	246.2	334.7
39	24.79	58.16	89	66.58	116.3	139	110.5	172.4	189	155.5	227.4	239	201.1	281.8	289	247.1	335.7
40	25.59	59.36	90	67.44	117.4	140	111.4	173.5	190	156.4	228.5	240	202.0	282.9	290	248.0	336.8
41	26.38	60.56	91	68.31	118.6	141	112.3	174.6	191	157.3	229.6	241	202.9	283.9	291	248.9	337.9
42	27.18	61.76	92	69.17	119.7	142	113.2	175.7	192	158.2	230.7	242	203.8	285.0	292	249.9	339.0
43	27.99	62.96	93	70.04	120.8	143	114.1	176.8	193	159.1	231.8	243	204.7	286.1	293	250.8	340.0
44	28.79	64.15	94	70.91	122.0	144	115.0	177.9	194	160.0	232.8	244	205.6	287.2	294	251.7	341.1
45	29.60	65.34	95	71.77	123.1	145	115.9	179.0	195	160.9	233.9	245	206.6	288.3	295	252.6	342.2
46	30.41	66.53	96	72.64	124.2	146	116.8	180.1	196	161.8	235.0	246	207.5	289.4	296	253.6	343.3
47	31.22	67.72	97	73.51	125.4	147	117.6	181.2	197	162.7	236.1	247	208.4	290.4	297	254.5	344.3
48	32.03	68.90	98	74.38	126.5	148	118.5	182.3	198	163.6	237.2	248	209.3	291.5	298	255.4	345.4
49	32.85	70.08	99	75.25	127.6	149	119.4	183.4	199	164.5	238.3	249	210.2	292.6	299	256.3	346.5

Table 13-14. *Lower (P⁻) and Upper (P⁺) 95% Confidence Limits for a Binomial Proportion, P (P=X/N)*

	X=0		X=1		X=2		X=3		X=4		X=5		X=6		X=7		X=8		X=9	
N	P⁻	P⁺	P⁻	P⁺	P⁻	P⁺	P⁻	P⁺	P⁻	P⁺	P⁻	P⁺	P⁻	P⁺	P⁻	P⁺	P⁻	P⁺	P⁻	P⁺
2	0	.842	013	.987	.158	1.00														
3	0	.708	.008	.906	.094	.992	.292	1.00												
4	0	.602	.006	.806	.068	.932	.194	.994	.398	1.00										
5	0	.522	.005	.716	.053	.853	.147	.947	.284	.995	.478	1.00								
6	0	.459	.004	.641	.043	.777	.118	.882	.223	.957	.359	.996	.541	1.00						
7	0	.410	.004	.579	.037	.710	.099	.816	.184	.901	.290	.963	.421	.996	.590	1.00				
8	0	.369	.003	.527	.032	.651	.085	.755	.157	.843	.245	.915	.349	.968	.473	.997	.631	1.00		
9	0	.336	.003	.482	.028	.600	.075	.701	.137	.788	.212	.863	.299	.925	.400	.972	.518	.997	664	1.00
10	0	.308	.003	.445	.025	.556	.067	.652	.122	.738	.187	.813	.262	.878	.348	.933	.444	.975	.555	.997
11	0	.285	.002	.413	.023	.518	.060	.610	.109	.692	.167	.766	.234	.833	.308	.891	.390	.940	.482	.977
12	0	.265	.002	.385	.021	.484	.055	.572	.099	.651	.152	.723	.211	.789	.277	.848	.349	.901	.428	.945
13	0	.247	.002	.360	.019	.454	.050	.538	.091	.614	.139	.684	.192	.749	.251	.808	.316	.861	.386	.909
14	0	.232	.002	.339	.018	.428	.047	.508	.084	.581	.128	.649	.177	.711	.230	.770	.289	.823	.351	.872
15	0	.218	.002	.319	.017	.405	.043	.481	.078	.551	.118	.616	.163	.677	.213	.734	.266	.787	.323	.837
16	0	.206	.002	.302	.016	.383	.040	.456	.073	.524	.110	.587	.152	.646	.198	.701	.247	.753	.299	.802
17	0	.195	.001	.287	.015	.364	.038	.434	.068	.499	.103	.560	.142	.617	.184	.671	.230	.722	.278	.770
18	0	.185	.001	.273	.014	.347	.036	.414	.064	.476	.097	.535	.133	.590	.173	.643	.215	.692	.260	.740
19	0	.176	.001	.260	.013	.331	.034	.396	.061	.456	.091	.512	.126	.566	.163	.616	.203	.665	.244	.711
20	0	.168	.001	.249	.012	.317	.032	.379	.057	.437	.087	.491	.119	.543	.154	.592	.191	.639	.231	.685
21	0	.161	.001	.238	.012	.304	.030	.363	.054	.419	.082	.472	.113	.522	.146	.570	.181	.616	.218	.660
22	0	.154	.001	.228	.011	.292	.029	.349	.052	.403	.078	.454	.107	.502	.139	.549	.172	.593	.207	.636
23	0	.148	.001	.219	.011	.280	.028	.336	.050	.388	.075	.437	.102	.484	.132	.529	.164	.573	.197	.615
24	0	.142	.001	.211	.010	.270	.027	.324	.047	.374	.071	.422	.098	.467	.126	.511	.156	.553	.188	.594
25	0	.137	.001	.204	.010	.260	.025	.312	.045	.361	.068	.407	.094	.451	.121	.494	.149	.535	.180	.575
26	0	.132	.001	.196	.009	.251	.024	.302	.044	.349	.066	.394	.090	.436	.116	.478	.143	.518	.172	.557
27	0	.128	.001	.190	.009	.243	.024	.292	.042	.337	.063	.381	.086	.423	.111	.463	.138	.502	.165	.540
28	0	.123	.001	.183	.009	.235	.023	.282	.040	.327	.061	.369	.083	.410	.107	.449	.132	.487	.159	.524
29	0	.119	.001	.178	.008	.228	.022	.274	.039	.317	.058	.358	.080	.397	.103	.435	.127	.472	.153	.508
30	0	.116	.001	.172	.008	.221	.021	.265	.038	.307	.056	.347	.077	.386	.099	.423	.123	.459	.147	.494
31	0	.112	.001	.167	.008	.214	.020	.258	.036	.298	.055	.337	.075	.375	.096	.411	.119	.446	.142	.480
32	0	.109	.001	.162	.008	.208	.020	.250	.035	.290	.053	.328	.072	.364	.093	.400	.115	.434	.137	.467
33	0	.106	.001	.158	.007	.202	.019	.243	.034	.282	.051	.319	.070	.355	.090	.389	.111	.423	.133	.455
34	0	.103	.001	.153	.007	.197	.019	.237	.033	.275	.050	.311	.068	.345	.087	.379	.107	.412	.129	.444
35	0	.100	.001	.149	.007	.192	.018	.231	.032	.267	.048	.303	.066	.336	.084	.369	.104	.401	.125	.433
36	0	.097	.001	.145	.007	.187	.018	.225	.031	.261	.047	.295	.064	.328	.082	.360	.101	.392	.121	.422
37	0	.095	.001	.142	.007	.182	.017	.219	.030	.254	.045	.288	.062	.320	.080	.352	.098	.382	.118	.412
38	0	.093	.001	.138	.006	.177	.017	.214	.029	.248	.044	.281	.060	.313	.077	.343	.096	.373	.114	.402
39	0	.090	.001	.135	.006	.173	.016	.209	.029	.242	.043	.274	.059	.305	.075	.335	.093	.365	.111	.393
40	0	.088	.001	.132	.006	.169	.016	.204	.028	.237	.042	.268	.057	.298	.073	.328	.091	.356	.108	.385
41	0	.086	.001	.129	.006	.165	.015	.199	.027	.231	.041	.262	.056	.292	.072	.321	.088	.349	.106	.376
42	0	.084	.001	.126	.006	.162	.015	.195	.027	.226	.040	.256	.054	.285	.070	.314	.086	.341	.103	.368
43	0	.082	.001	.123	.006	.158	.015	.191	.026	.221	.039	.251	.053	.279	.068	.307	.084	.334	.100	.360
44	0	.080	.001	.120	.006	.155	.014	.187	.025	.217	.038	.246	.052	.274	.066	.301	.082	.327	.098	.353
45	0	.079	.001	.118	.005	.151	.014	.183	.025	.212	.037	.241	.051	.268	.065	.295	.080	.321	.096	.346
46	0	.077	.001	.115	.005	.148	.014	.179	.024	.208	.036	.236	.049	.263	.063	.289	.078	.314	.094	.339
47	0	.075	.001	.113	.005	.145	.013	.175	.024	.204	.035	.231	.048	.257	.062	.283	.076	.308	.091	.333
48	0	.074	.001	.111	.005	.143	.013	.172	.023	.200	.035	.227	.047	.252	.061	.278	.075	.302	.089	.326
49	0	.073	.001	.109	.005	.140	.013	.169	.023	.196	.034	.222	.046	.248	.059	.272	.073	.297	.088	.320
50	0	.071	.001	.106	.005	.137	.013	.165	.022	.192	.033	.218	.045	.243	.058	.267	.072	.291	.086	.314
51	0	.070	.000	.104	.005	.135	.012	.162	.022	.189	.033	.214	.044	.239	.057	.263	.070	.286	.084	.309
52	0	.068	.000	.103	.005	.132	.012	.159	.021	.185	.032	.210	.044	.234	.056	.258	.069	.281	.082	.303
53	0	.067	.000	.101	.005	.130	.012	.157	.021	.182	.031	.207	.043	.230	.055	.253	.067	.276	.081	.298
54	0	.066	.000	.099	.005	.127	.012	.154	.021	.179	.031	.203	.042	.226	.054	.249	.066	.271	.079	.293
55	0	.065	.000	.097	.004	.125	.011	.151	.020	.176	.030	.200	.041	.222	.053	.245	.065	.267	.078	.288
56	0	.064	.000	.096	.004	.123	.011	.149	.020	.173	.030	.196	.040	.219	.052	.241	.064	.262	.076	.283
57	0	.063	.000	.094	.004	.121	.011	.146	.019	.170	.029	.193	.040	.215	.051	.237	.063	.258	.075	.279
58	0	.062	.000	.092	.004	.119	.011	.144	.019	.167	.029	.190	.039	.212	.050	.233	.061	.254	.073	.274
59	0	.061	.000	.091	.004	.117	.011	.141	.019	.165	.028	.187	.038	.208	.049	.229	.060	.250	.072	.270
60	0	.060	.000	.089	.004	.115	.010	.139	.018	.162	.028	.184	.038	.205	.048	.226	.059	.246	.071	.266

(continued)

Table 13-14. *(continued)*

N	X=0 P-	P+	X=1 P-	P+	X=2 P-	P+	X=3 P-	P+	X=4 P-	P+	X=5 P-	P+	X=6 P-	P+	X=7 P-	P+	X=8 P-	P+	X=9 P-	P+
10	692	1.00																		
11	.587	.998	.715	1.00																
12	.516	.979	.615	.998	.735	1.00														
13	.462	.950	.546	.981	.640	.998	.753	1.00												
14	.419	.916	.492	.953	.572	.982	.661	.998	.768	1.00										
15	.384	.882	.449	.922	.519	.957	.595	.983	.681	.998	.782	1.00								
16	.354	.848	.413	.890	.476	.927	.544	.960	.617	.984	.698	.998	.794	1.00						
17	.329	.816	.383	.858	.440	.897	.501	.932	.566	.962	.636	.985	.713	.999	.805	1.00				
18	.308	.785	.357	.827	.410	.867	.465	.903	.524	.936	.586	.964	.653	.986	.727	.999	815	1.00		
19	.289	.756	.335	.797	.384	.837	.434	.874	.488	.909	.544	.939	.604	.966	.669	.987	.740	.999	.824	1.00
20	.272	.728	.315	.769	.361	.809	.408	.846	.457	.881	.509	.913	.563	.943	.621	.968	.683	.988	.751	.999
21	.257	.702	.298	.743	.340	.782	.384	.819	.430	.854	.478	.887	.528	.918	.581	.946	.637	.970	.696	.988
22	.244	.678	.282	.718	.322	.756	.364	.793	.407	.828	.451	.861	.498	.893	.546	.922	.597	.948	.651	.971
23	.232	.655	.268	.694	.306	.732	.345	.768	.385	.803	.427	.836	.471	.868	.516	.898	.563	.925	.612	.950
24	.221	.634	.256	.672	.291	.709	.328	.744	.366	.779	.406	.812	.447	.844	.489	.874	.533	.902	.578	.929
25	.211	.613	.244	.651	.278	.687	.313	.722	.349	.756	.387	.789	.425	.820	.465	.851	.506	.879	.549	.906
26	.202	.594	.234	.631	.266	.666	.299	.701	.334	.734	.369	.766	.406	.798	.443	.828	.482	.857	.522	.884
27	.194	.576	.224	.612	.255	.647	.287	.681	.319	.713	.353	.745	.388	.776	.424	.806	.460	.835	.498	.862
28	.186	.559	.215	.594	.245	.628	.275	.661	.306	.694	.339	.725	.372	.755	.406	.785	.441	.814	.476	.841
29	.179	.543	.207	.577	.235	.611	.264	.643	.294	.675	.325	.706	.357	.736	.389	.765	.423	.793	.457	.821
30	.173	.528	.199	.561	.227	.594	.255	.626	.283	.657	.313	.687	.343	.717	.374	.745	.406	.773	.439	.801
31	.167	.514	.192	.546	.218	.578	.245	.609	.273	.640	.302	.669	.331	.698	.360	.727	.391	.755	.422	.782
32	.161	.500	.186	.532	.211	.563	.237	.594	.264	.623	.291	.653	.319	.681	.347	.709	.377	.736	.406	.763
33	.156	.487	.180	.518	.204	.549	.229	.579	.255	.608	.281	.636	.308	.665	.335	.692	.364	.719	.392	.745
34	.151	.475	.174	.505	.197	.535	.222	.564	.246	.593	.272	.621	.298	.649	.324	.676	.351	.702	.379	.728
35	.146	.463	.169	.493	.191	.522	.215	.551	.239	.579	.263	.606	.288	.634	.314	.660	.340	.686	.366	.712
36	.142	.452	.163	.481	.186	.510	.208	.538	.231	.565	.255	.592	.279	.619	.304	.645	.329	.671	.355	.696
37	.138	.441	.159	.470	.180	.498	.202	.525	.225	.552	.248	.579	.271	.605	.295	.631	.319	.656	.344	.681
38	.134	.431	.154	.459	.175	.487	.196	.514	.218	.540	.240	.566	.263	.592	.286	.617	.310	.642	.334	.666
39	.130	.421	.150	.449	.170	.476	.191	.502	.212	.528	.234	.554	.256	.579	.278	.604	.301	.628	.324	.652
40	.127	.412	.146	.439	.166	.465	.186	.491	.206	.517	.227	.542	.249	.567	.270	.591	.293	.615	.315	.639
41	.124	.403	.142	.429	.161	.455	.181	.481	.201	.506	.221	.531	.242	.555	.263	.579	.285	.603	.307	.626
42	.121	.395	.139	.420	.157	.446	.176	.471	.196	.495	.216	.520	.236	.544	.256	.567	.277	.590	.298	.613
43	.118	.386	.135	.412	.153	.437	.172	.461	.191	.485	.210	.509	.230	.533	.250	.556	.270	.579	.291	.601
44	.115	.378	.132	.403	.150	.428	.168	.452	.186	.476	.205	.499	.224	.522	.244	.545	.263	.568	.283	.590
45	.112	.371	.129	.395	.146	.419	.164	.443	.182	.466	.200	.490	.219	.512	.238	.535	.257	.557	.277	.578
46	.109	.364	.126	.388	.143	.411	.160	.435	.177	.458	.195	.480	.214	.502	.232	.525	.251	.546	.270	.568
47	.107	.357	.123	.380	.139	.403	.156	.426	.173	.449	.191	.471	.209	.493	.227	.515	.245	.536	.264	.557
48	.105	.350	.120	.373	.136	.396	.153	.418	.170	.441	.187	.463	.204	.484	.222	.505	.240	.526	.258	.547
49	.102	.343	.118	.366	.133	.389	.149	.411	.166	.433	.183	.454	.199	.475	.217	.496	.234	.517	.252	.538
50	.100	.337	.115	.360	.131	.382	.146	.403	.162	.425	.179	.446	.195	.467	.212	.488	.229	.508	.247	.528
51	.098	.331	.113	.353	.128	.375	.143	.396	.159	.417	.175	.438	.191	.459	.208	.479	.224	.499	.241	.519
52	.096	.325	.111	.347	.125	.368	.140	.389	.156	.410	.171	.431	.187	.451	.203	.471	.220	.491	.236	.510
53	.094	.320	.108	.341	.123	.362	.138	.383	.153	.403	.168	.423	.183	.443	.199	.463	.215	.483	.231	.502
54	.093	.314	.106	.335	.120	.356	.135	.376	.150	.397	.165	.416	.180	.436	.195	.456	.211	.475	.227	.494
55	.091	.309	.104	.330	.118	.350	.132	.370	.147	.390	.161	.410	.176	.429	.191	.448	.207	.467	.222	.486
56	.089	.304	.102	.324	.116	.344	.130	.364	.144	.384	.158	.403	.173	.422	.188	.441	.203	.460	.218	.478
57	.087	.299	.100	.319	.114	.339	.127	.358	.141	.378	.155	.397	.170	.415	.184	.434	.199	.452	.214	.471
58	.086	.294	.099	.314	.112	.334	.125	.353	.139	.372	.153	.390	.167	.409	.181	.427	.195	.445	.210	.463
59	.084	.290	.097	.309	.110	.328	.123	.347	.136	.366	.150	.384	.164	.403	.178	.421	.192	.439	.206	.456
60	.083	.285	.095	.304	.108	.323	.121	.342	.134	.360	.147	.379	.161	.397	.175	.414	.188	.432	.203	.450

N = number of trials, X = number of "successes."
For P>0.5, use confidence limits based on 1-P.

Table 13-15. *Lower (P⁻) and Upper (P⁺) Simultaneous 95% Confidence Limits for Multinomial Proportions (P)*

 # Groups = 3

	P=0.05		P=.10		P=0.15		P=0.20		P=0.25		P=0.30		P=0.35		P=0.40		P=0.45		P=0.50	
N	P⁻	P⁺	P⁻	P⁺	P⁻	P⁺	P⁻	P⁺	P⁻	P⁺	P⁻	P⁺	P⁻	P⁺	P⁻	P⁺	P⁻	P⁺	P⁻	P⁺
20	.000	.296	.007	.366	.021	.429	.041	.486	066	.540	.094	.590	.125	.638	.159	.683	.195	.726	.264	.736
21	.001	.288	.008	.358	.023	.420	.044	.478	.069	.532	.098	.583	.129	.631	.163	.677	.200	.720	.268	.732
22	.001	.280	.008	.350	.024	.413	.046	.471	.072	.525	.101	.576	.133	.624	.168	.670	.234	.687	.273	.727
23	.001	.273	.009	.343	.026	.406	.048	.464	.074	.518	.104	.569	.137	.618	.172	.664	.238	.682	.277	.723
24	.001	.266	.010	.336	.027	.399	.050	.457	.077	.512	.107	.563	.141	.612	.176	.659	.241	.678	.280	.720
25	.001	.259	.011	.330	.028	.393	.052	.451	.079	.506	.110	.558	.144	.607	.206	.631	.244	.674	.284	.716
26	.001	.253	.011	.324	.030	.387	.054	.445	.082	.500	.113	.552	.147	.602	.209	.627	.247	.671	.288	.712
27	.001	.248	.012	.318	.031	.382	.056	.440	.084	.495	.116	.547	.150	.597	.212	.623	.250	.667	.291	.709
28	.001	.243	.013	.313	.032	.376	.057	.435	.086	.490	.119	.542	.153	.592	.214	.620	.253	.664	.294	.706
29	.002	.238	.013	.308	.033	.372	.059	.430	.088	.486	.121	.538	.179	.570	.217	.616	.256	.661	.297	.703
30	.002	.233	.014	.304	.035	.367	.061	.426	.090	.481	.123	.534	.182	.567	.219	.613	.259	.657	.300	.700
31	.002	.229	.015	.299	.036	.363	.062	.422	.092	.477	.126	.529	.184	.563	.221	.610	.261	.654	.302	.698
32	.002	.225	.015	.295	.037	.359	.064	.417	.094	.473	.128	.526	.186	.560	.224	.607	.264	.652	.305	.695
33	.002	.221	.016	.291	.038	.355	.065	.414	.096	.469	.130	.522	.187	.557	.226	.604	.266	.649	.308	.692
34	.002	.217	.017	.288	.039	.351	.067	.410	.098	.465	.152	.505	.189	.554	.228	.601	.268	.646	.310	.690
35	.003	.214	.017	.284	.040	.347	.068	.406	.100	.462	.154	.502	.191	.551	.230	.598	.270	.644	.312	.688
36	.003	.211	.018	.281	.041	.344	.069	.403	.101	.459	.155	.499	.193	.549	.232	.596	.272	.641	.315	.685
37	.003	.207	.018	.278	.042	.341	.071	.400	.103	.455	.157	.497	.194	.546	.233	.593	.274	.639	.317	.683
38	.003	.204	.019	.274	.043	.338	.072	.397	.104	.452	.158	.494	.196	.543	.235	.591	.276	.637	.319	.681
39	.003	.202	.020	.272	.044	.335	.073	.394	.106	.449	.160	.492	.197	.541	.237	.589	.278	.635	.321	.679
40	.003	.199	.020	.269	.045	.332	.074	.391	.107	.447	.161	.489	.199	.539	.239	.586	.280	.632	.323	.677
41	.004	.196	.021	.266	.046	.329	.076	.388	.126	.435	.162	.487	.200	.536	.240	.584	.282	.630	.325	.675
42	.004	.194	.021	.263	.047	.327	.077	.385	.127	.433	.163	.485	.202	.534	.242	.582	.284	.628	.327	.673
43	.004	.191	.022	.261	.047	.324	.078	.383	.128	.431	.165	.482	.203	.532	.243	.580	.285	.627	.329	.671
44	.004	.189	.022	.259	.048	.322	.079	.381	.129	.429	.166	.480	.204	.530	.245	.578	.287	.625	.330	.670
45	.004	.187	.023	.256	.049	.319	.080	.378	.130	.426	.167	.478	.206	.528	.246	.576	.288	.623	.332	.668
46	.004	.185	.023	.254	.050	.317	.081	.376	.131	.424	.168	.476	.207	.526	.248	.575	.290	.621	.334	.666
47	.005	.182	.024	.252	.050	.315	.082	.374	.132	.422	.169	.474	.208	.524	.249	.573	.291	.620	.335	.665
48	.005	.181	.024	.250	.051	.313	.083	.372	.133	.421	.170	.473	.209	.523	.250	.571	.293	.618	.337	.663
49	.005	.179	.025	.248	.052	.311	.084	.370	.134	.419	.171	.471	.211	.521	.252	.569	.294	.616	.338	.662
50	.005	.177	.025	.246	.053	.309	.085	.368	.134	.417	.172	.469	.212	.519	.253	.568	.296	.615	.340	.660
60	.007	.161	.029	.230	.059	.292	.105	.347	.142	.402	.181	.454	.222	.505	.264	.554	.307	.601	.352	.648
70	.008	.150	.033	.218	.075	.278	.110	.335	.148	.390	.188	.442	.230	.493	.273	.543	.317	.591	.362	.638
80	.010	.141	.036	.208	.078	.269	.115	.325	.154	.380	.194	.433	.236	.484	.280	.534	.325	.582	.371	.629
90	.011	.134	.039	.201	.081	.261	.118	.317	.158	.372	.199	.425	.242	.476	.286	.526	.331	.575	.378	.622
100	.012	.128	.041	.194	.084	.254	.122	.311	.162	.365	.204	.418	.247	.469	.291	.520	.337	.569	.384	.616
110	.013	.123	.050	.189	.086	.249	.125	.305	.165	.360	.207	.412	.251	.464	.296	.514	.342	.563	.389	.611
120	.014	.119	.052	.185	.088	.244	.127	.300	.168	.355	.211	.407	.255	.459	.300	.509	.346	.559	.393	.607
130	.015	.116	.053	.181	.090	.240	.130	.296	.171	.350	.214	.403	.258	.455	.303	.505	.350	.554	.397	.603
140	.016	.113	.054	.177	.092	.236	.132	.292	.173	.346	.217	.399	.261	.451	.307	.501	.353	.551	.401	.599
150	.017	.110	.055	.174	.093	.233	.134	.289	.176	.343	.219	.396	.264	.447	.310	.498	.356	.547	.404	.596
160	.018	.107	.056	.171	.095	.230	.135	.285	.178	.340	.221	.392	.266	.444	.312	.495	.359	.544	.407	.593
170	.019	.105	.057	.169	.096	.227	.137	.283	.180	.337	.224	.390	.269	.441	.315	.492	.362	.541	.410	.590
180	.019	.103	.058	.166	.097	.224	.138	.280	.181	.334	.225	.387	.271	.439	.317	.489	.364	.539	.412	.588
190	.020	.101	.059	.164	.098	.222	.140	.278	.183	.332	.227	.384	.273	.436	.319	.487	.366	.537	.414	.586
200	.020	.100	.060	.162	.099	.220	.141	.276	.184	.330	.229	.382	.274	.434	.321	.485	.368	.534	.417	.583
250	.026	.094	.063	.155	.104	.212	.146	.267	.191	.321	.236	.373	.282	.425	.329	.476	.377	.526	.425	.575
300	.027	.089	.066	.149	.107	.206	.151	.261	.195	.314	.241	.367	.287	.418	.335	.469	.383	.519	.432	.568
350	.029	.086	.068	.145	.110	.201	.154	.256	.199	.309	.245	.361	.292	.413	.339	.464	.388	.514	.437	.563
400	.030	.083	.070	.142	.112	.198	.157	.252	.202	.305	.248	.357	.295	.409	.343	.460	.392	.510	.441	.559
450	.031	.081	.071	.139	.114	.195	.159	.249	.204	.302	.251	.354	.298	.405	.346	.456	.395	.506	.444	.556
500	.031	.079	.072	.137	.116	.192	.161	.246	.207	.299	.253	.351	.301	.403	.349	.453	.398	.504	.447	.553
1000	.036	.069	.080	.125	.125	.179	.171	.232	.219	.284	.267	.336	.315	.387	.364	.438	.413	.488	.462	.538

(continued)

Table 13-15. *(continued)*

 # Groups = 4

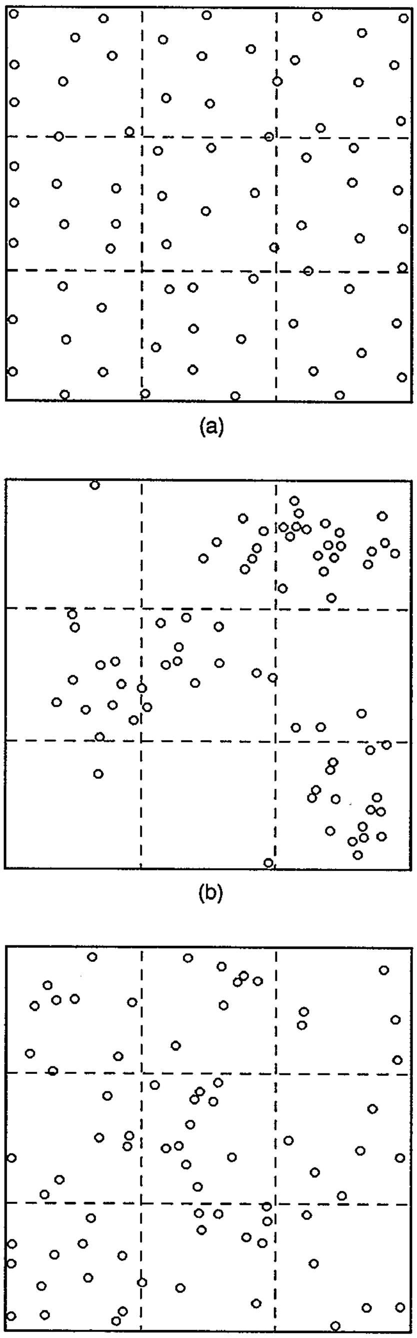
N	P=0.05 P-	P=0.05 P+	P=.10 P-	P=.10 P+	P=0.15 P-	P=0.15 P+	P=0.20 P-	P=0.20 P+	P=0.25 P-	P=0.25 P+	P=0.30 P-	P=0.30 P+	P=0.35 P-	P=0.35 P+	P=0.40 P-	P=0.40 P+	P=0.45 P-	P=0.45 P+	P=0.50 P-	P=0.50 P+
20	.000	.308	.006	.378	.019	.440	.038	.498	.061	.551	.088	.601	.119	.649	.151	.693	.187	.736	.256	.744
21	.000	.299	.007	.369	.021	.432	.040	.490	.064	.543	.092	.594	.123	.641	.156	.687	.192	.729	.261	.739
22	.000	.291	.007	.361	.022	.424	.042	.482	.067	.536	.096	.587	.127	.635	.161	.680	.197	.723	.265	.735
23	.001	.283	.008	.354	.023	.417	.044	.475	.070	.529	.099	.580	.131	.628	.165	.674	.231	.691	.269	.731
24	.001	.276	.009	.347	.025	.410	.046	.468	.072	.522	.102	.574	.134	.622	.169	.669	.234	.687	.273	.727
25	.001	.270	.009	.340	.026	.403	.048	.462	.075	.516	.105	.568	.138	.617	.200	.640	.237	.683	.277	.723
26	.001	.263	.010	.334	.027	.397	.050	.456	.077	.511	.108	.562	.141	.611	.203	.636	.241	.679	.280	.720
27	.001	.258	.011	.328	.029	.392	.052	.450	.080	.505	.111	.557	.144	.606	.206	.632	.244	.675	.283	.717
28	.001	.252	.011	.323	.030	.386	.054	.445	.082	.500	.113	.552	.147	.602	.208	.628	.247	.672	.287	.713
29	.001	.247	.012	.318	.031	.381	.055	.440	.084	.495	.116	.547	.174	.579	.211	.625	.249	.668	.290	.710
30	.001	.242	.013	.313	.032	.377	.057	.435	.086	.491	.118	.543	.176	.575	.213	.621	.252	.665	.293	.707
31	.002	.238	.013	.309	.033	.372	.059	.431	.088	.486	.120	.539	.178	.572	.216	.618	.255	.662	.295	.705
32	.002	.233	.014	.304	.034	.368	.060	.427	.090	.482	.123	.535	.180	.569	.218	.615	.257	.659	.298	.702
33	.002	.229	.014	.300	.035	.364	.062	.423	.092	.478	.125	.531	.182	.566	.220	.612	.259	.656	.301	.699
34	.002	.226	.015	.296	.036	.360	.063	.419	.093	.474	.148	.514	.184	.562	.222	.609	.262	.654	.303	.697
35	.002	.222	.016	.293	.037	.356	.064	.415	.095	.471	.149	.511	.186	.560	.224	.606	.264	.651	.306	.694
36	.002	.218	.016	.289	.038	.353	.066	.412	.097	.467	.151	.508	.188	.557	.226	.604	.266	.649	.308	.692
37	.002	.215	.017	.286	.039	.349	.067	.408	.098	.464	.152	.505	.189	.554	.228	.601	.268	.646	.310	.690
38	.003	.212	.017	.283	.040	.346	.068	.405	.100	.461	.154	.503	.191	.552	.230	.599	.270	.644	.312	.688
39	.003	.209	.018	.280	.041	.343	.070	.402	.102	.458	.155	.500	.192	.549	.231	.596	.272	.642	.314	.686
40	.003	.206	.018	.277	.042	.340	.071	.399	.103	.455	.157	.497	.194	.547	.233	.594	.274	.640	.316	.684
41	.003	.203	.019	.274	.043	.337	.072	.396	.122	.444	.158	.495	.195	.544	.235	.592	.276	.638	.318	.682
42	.003	.201	.020	.271	.044	.335	.073	.394	.123	.441	.159	.493	.197	.542	.236	.590	.277	.635	.320	.680
43	.003	.198	.020	.269	.045	.332	.074	.391	.124	.439	.160	.490	.198	.540	.238	.588	.279	.634	.322	.678
44	.004	.196	.021	.266	.045	.329	.075	.388	.125	.437	.161	.488	.200	.538	.239	.585	.281	.632	.324	.676
45	.004	.193	.021	.264	.046	.327	.076	.386	.126	.435	.163	.486	.201	.536	.241	.584	.282	.630	.326	.674
46	.004	.191	.022	.261	.047	.325	.077	.384	.127	.432	.164	.484	.202	.534	.242	.582	.284	.628	.327	.673
47	.004	.189	.022	.259	.048	.322	.078	.381	.128	.430	.165	.482	.203	.532	.244	.580	.285	.626	.329	.671
48	.004	.187	.023	.257	.049	.320	.079	.379	.129	.428	.166	.480	.205	.530	.245	.578	.287	.625	.330	.670
49	.004	.185	.023	.255	.049	.318	.080	.377	.130	.426	.167	.478	.206	.528	.246	.576	.288	.623	.332	.668
50	.005	.183	.023	.253	.050	.316	.081	.375	.131	.425	.168	.476	.207	.526	.248	.575	.290	.621	.333	.667
60	.006	.167	.028	.236	.056	.299	.102	.354	.139	.409	.177	.461	.217	.511	.259	.560	.302	.607	.347	.653
70	.008	.155	.031	.223	.073	.285	.108	.342	.145	.396	.184	.448	.225	.499	.268	.549	.312	.596	.357	.643
80	.009	.146	.034	.213	.076	.275	.112	.331	.150	.386	.190	.439	.232	.490	.275	.539	.320	.587	.366	.634
90	.010	.138	.037	.205	.079	.266	.116	.323	.155	.378	.196	.430	.238	.482	.282	.531	.327	.580	.373	.627
100	.012	.132	.039	.199	.082	.259	.119	.316	.159	.371	.200	.423	.243	.475	.287	.525	.332	.574	.379	.621
110	.013	.127	.049	.194	.084	.254	.122	.310	.162	.365	.204	.417	.247	.469	.292	.519	.337	.568	.384	.616
120	.014	.123	.050	.189	.086	.249	.125	.305	.165	.359	.208	.412	.251	.464	.296	.514	.342	.563	.389	.611
130	.015	.119	.052	.185	.088	.244	.127	.300	.168	.355	.211	.408	.255	.459	.300	.510	.346	.559	.393	.607
140	.015	.116	.053	.181	.090	.240	.129	.296	.171	.351	.213	.404	.258	.455	.303	.506	.349	.555	.397	.603
150	.016	.113	.054	.178	.091	.237	.131	.293	.173	.347	.216	.400	.260	.451	.306	.502	.353	.551	.400	.600
160	.017	.110	.055	.175	.093	.234	.133	.290	.175	.344	.218	.397	.263	.448	.309	.499	.355	.548	.403	.597
170	.018	.108	.056	.172	.094	.231	.135	.287	.177	.341	.221	.394	.265	.445	.311	.496	.358	.545	.406	.594
180	.018	.106	.057	.170	.095	.228	.136	.284	.179	.338	.223	.391	.268	.442	.314	.493	.361	.543	.408	.592
190	.019	.104	.058	.168	.096	.226	.138	.281	.180	.336	.224	.388	.270	.440	.316	.491	.363	.540	.411	.589
200	.019	.102	.059	.166	.098	.224	.139	.279	.182	.333	.226	.386	.271	.438	.318	.488	.365	.538	.413	.587
250	.025	.097	.062	.158	.102	.215	.144	.270	.188	.324	.233	.377	.279	.428	.326	.479	.374	.529	.422	.578
300	.027	.092	.065	.152	.106	.209	.149	.264	.193	.317	.239	.370	.285	.421	.332	.472	.380	.522	.429	.571
350	.028	.088	.067	.147	.108	.204	.152	.258	.197	.312	.243	.364	.289	.416	.337	.467	.385	.517	.434	.566
400	.029	.085	.068	.144	.111	.200	.155	.254	.200	.308	.246	.360	.293	.411	.341	.462	.389	.512	.438	.562
450	.030	.082	.070	.141	.113	.197	.157	.251	.203	.304	.249	.356	.296	.408	.344	.459	.393	.509	.442	.558
500	.031	.080	.071	.139	.114	.194	.159	.248	.205	.301	.252	.353	.299	.405	.347	.456	.395	.506	.444	.556
1000	.035	.070	.079	.126	.124	.180	.170	.233	.217	.286	.265	.337	.313	.388	.362	.439	.411	.489	.461	.539

(continued)

Table 13-15. *(continued)*

 # Groups = 5

	P=0.05		P=.10		P=0.15		P=0.20		P=0.25		P=0.30		P=0.35		P=0.40		P=0.45		P=0.50	
N	P-	P+	P-	P+	P-	P+	P-	P+	P-	P+	P-	P+	P-	P+	P-	P+	P-	P+	P-	P+
20	.000	.317	.005	.387	.018	.449	.036	.507	.058	.560	.085	.610	.114	.657	.146	.701	.181	.743	.250	.750
21	.000	.308	.006	.378	.019	.441	.038	.498	.061	.552	.088	.602	.118	.649	.151	.694	.186	.736	.255	.745
22	.000	.299	.007	.370	.020	.433	.040	.490	.064	.544	.092	.595	.122	.642	.155	.687	.191	.730	.259	.741
23	.000	.291	.007	.362	.022	.425	.042	.483	.067	.537	.095	.588	.126	.636	.160	.681	.226	.697	.263	.737
24	.001	.284	.008	.355	.023	.418	.044	.476	.069	.530	.098	.581	.130	.630	.164	.676	.229	.693	.267	.733
25	.001	.277	.008	.348	.024	.412	.046	.470	.072	.524	.101	.575	.133	.624	.168	.670	.232	.689	.271	.729
26	.001	.271	.009	.342	.026	.405	.048	.464	.074	.518	.104	.570	.136	.619	.199	.642	.236	.685	.275	.725
27	.001	.265	.010	.336	.027	.400	.049	.458	.076	.513	.107	.564	.140	.614	.201	.638	.239	.681	.278	.722
28	.001	.259	.010	.330	.028	.394	.051	.453	.079	.508	.109	.559	.143	.609	.204	.634	.242	.678	.281	.719
29	.001	.254	.011	.325	.029	.389	.053	.448	.081	.503	.112	.555	.146	.604	.206	.631	.244	.674	.284	.716
30	.001	.249	.012	.320	.030	.384	.054	.443	.083	.498	.114	.550	.172	.582	.209	.627	.247	.671	.287	.713
31	.001	.244	.012	.316	.031	.379	.056	.438	.085	.493	.117	.546	.174	.578	.211	.624	.250	.668	.290	.710
32	.001	.240	.013	.311	.032	.375	.058	.434	.087	.489	.119	.542	.176	.575	.213	.621	.252	.665	.293	.707
33	.002	.236	.013	.307	.034	.371	.059	.430	.088	.485	.121	.538	.178	.572	.216	.618	.255	.662	.295	.705
34	.002	.232	.014	.303	.035	.367	.060	.426	.090	.481	.123	.534	.180	.569	.218	.615	.257	.659	.298	.702
35	.002	.228	.015	.299	.036	.363	.062	.422	.092	.478	.146	.518	.182	.566	.220	.612	.259	.657	.300	.700
36	.002	.225	.015	.296	.037	.359	.063	.418	.094	.474	.148	.515	.184	.563	.222	.609	.261	.654	.303	.697
37	.002	.221	.016	.292	.037	.356	.064	.415	.095	.471	.149	.512	.185	.560	.224	.607	.263	.652	.305	.695
38	.002	.218	.016	.289	.038	.353	.066	.412	.097	.467	.151	.509	.187	.558	.225	.604	.265	.649	.307	.693
39	.002	.215	.017	.286	.039	.349	.067	.408	.098	.464	.152	.506	.189	.555	.227	.602	.267	.647	.309	.691
40	.003	.212	.017	.283	.040	.346	.068	.405	.100	.461	.153	.504	.190	.553	.229	.600	.269	.645	.311	.689
41	.003	.209	.018	.280	.041	.343	.069	.402	.101	.458	.155	.501	.192	.550	.231	.597	.271	.643	.313	.687
42	.003	.206	.018	.277	.042	.341	.070	.400	.121	.448	.156	.499	.193	.548	.232	.595	.273	.641	.315	.685
43	.003	.204	.019	.274	.043	.338	.072	.397	.122	.445	.157	.496	.195	.546	.234	.593	.275	.639	.317	.683
44	.003	.201	.019	.272	.044	.335	.073	.394	.123	.443	.158	.494	.196	.543	.235	.591	.276	.637	.319	.681
45	.003	.199	.020	.269	.044	.333	.074	.392	.124	.441	.159	.492	.197	.541	.237	.589	.278	.635	.321	.679
46	.004	.196	.020	.267	.045	.330	.075	.389	.125	.438	.161	.490	.199	.539	.238	.587	.280	.633	.322	.678
47	.004	.194	.021	.264	.046	.328	.076	.387	.126	.436	.162	.488	.200	.537	.240	.585	.281	.631	.324	.676
48	.004	.192	.021	.262	.047	.326	.077	.385	.126	.434	.163	.486	.201	.535	.241	.583	.283	.630	.326	.674
49	.004	.190	.022	.260	.047	.323	.078	.383	.127	.432	.164	.484	.202	.534	.242	.582	.284	.628	.327	.673
50	.004	.188	.022	.258	.048	.321	.079	.380	.128	.430	.165	.482	.203	.532	.244	.580	.285	.626	.329	.671
60	.006	.171	.026	.241	.054	.304	.100	.360	.136	.414	.174	.466	.214	.516	.255	.565	.298	.612	.342	.658
70	.007	.159	.030	.227	.060	.290	.105	.347	.142	.401	.181	.453	.222	.504	.264	.553	.308	.601	.353	.647
80	.009	.149	.033	.217	.074	.279	.110	.336	.148	.390	.188	.443	.229	.494	.272	.543	.316	.592	.362	.638
90	.010	.141	.036	.209	.077	.271	.114	.327	.152	.382	.193	.435	.235	.486	.278	.535	.323	.584	.369	.631
100	.011	.135	.038	.202	.080	.263	.117	.320	.156	.375	.197	.427	.240	.479	.284	.529	.329	.577	.375	.625
110	.012	.130	.048	.198	.082	.257	.120	.314	.160	.368	.201	.421	.244	.473	.289	.523	.334	.572	.381	.619
120	.013	.125	.049	.193	.085	.252	.123	.309	.163	.363	.205	.416	.248	.467	.293	.517	.339	.567	.386	.614
130	.014	.121	.051	.188	.086	.247	.125	.304	.166	.358	.208	.411	.252	.463	.297	.513	.343	.562	.390	.610
140	.015	.118	.052	.184	.088	.243	.127	.300	.169	.354	.211	.407	.255	.458	.300	.509	.346	.558	.394	.606
150	.016	.115	.053	.181	.090	.240	.129	.296	.171	.350	.214	.403	.258	.455	.303	.505	.350	.555	.397	.603
160	.016	.112	.054	.178	.091	.237	.131	.293	.173	.347	.216	.400	.261	.451	.306	.502	.353	.551	.400	.600
170	.017	.110	.055	.175	.093	.234	.133	.290	.175	.344	.218	.397	.263	.448	.309	.499	.355	.548	.403	.597
180	.018	.108	.056	.173	.094	.231	.134	.287	.177	.341	.220	.394	.265	.445	.311	.496	.358	.546	.406	.594
190	.018	.106	.057	.170	.095	.228	.136	.284	.178	.338	.222	.391	.267	.443	.313	.493	.360	.543	.408	.592
200	.019	.104	.058	.168	.096	.226	.137	.282	.180	.336	.224	.389	.269	.440	.315	.491	.362	.541	.410	.590
250	.025	.099	.061	.160	.101	.217	.143	.273	.187	.326	.231	.379	.277	.431	.324	.481	.371	.531	.420	.580
300	.026	.093	.064	.154	.105	.211	.147	.266	.191	.319	.237	.372	.283	.423	.330	.474	.378	.524	.426	.574
350	.027	.089	.066	.149	.107	.206	.151	.260	.195	.314	.241	.366	.288	.418	.335	.469	.383	.519	.432	.568
400	.029	.086	.068	.145	.110	.202	.154	.256	.199	.310	.245	.362	.291	.413	.339	.464	.387	.514	.436	.564
450	.029	.084	.069	.142	.112	.198	.156	.253	.201	.306	.248	.358	.295	.410	.342	.461	.391	.511	.440	.560
500	.030	.082	.071	.140	.113	.196	.158	.250	.204	.303	.250	.355	.297	.407	.345	.457	.394	.508	.443	.557
1000	.035	.071	.078	.127	.123	.181	.169	.235	.216	.287	.264	.339	.312	.390	.361	.440	.410	.491	.459	.541


(a)

(b)

(c)

Figure 13-1. Dispersion of PFU in suspension. Dashed lines represent separate cell culture bottles. Types of dispersion represented are (a) uniform, (b) clustered, and (c) random (Poisson).

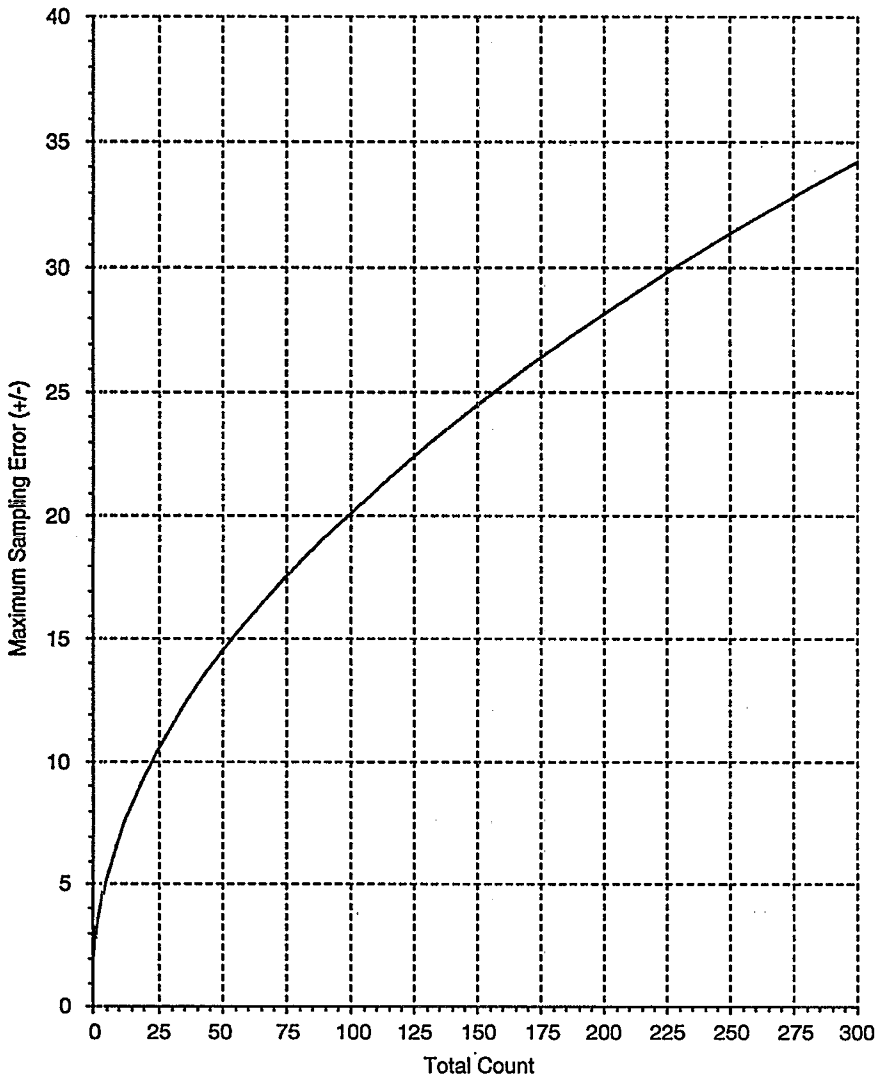


Figure 13-2. Maximum sampling error of total plaque count half-length of 95% confidence interval, assuming randomly dispersed (Poisson) counts.

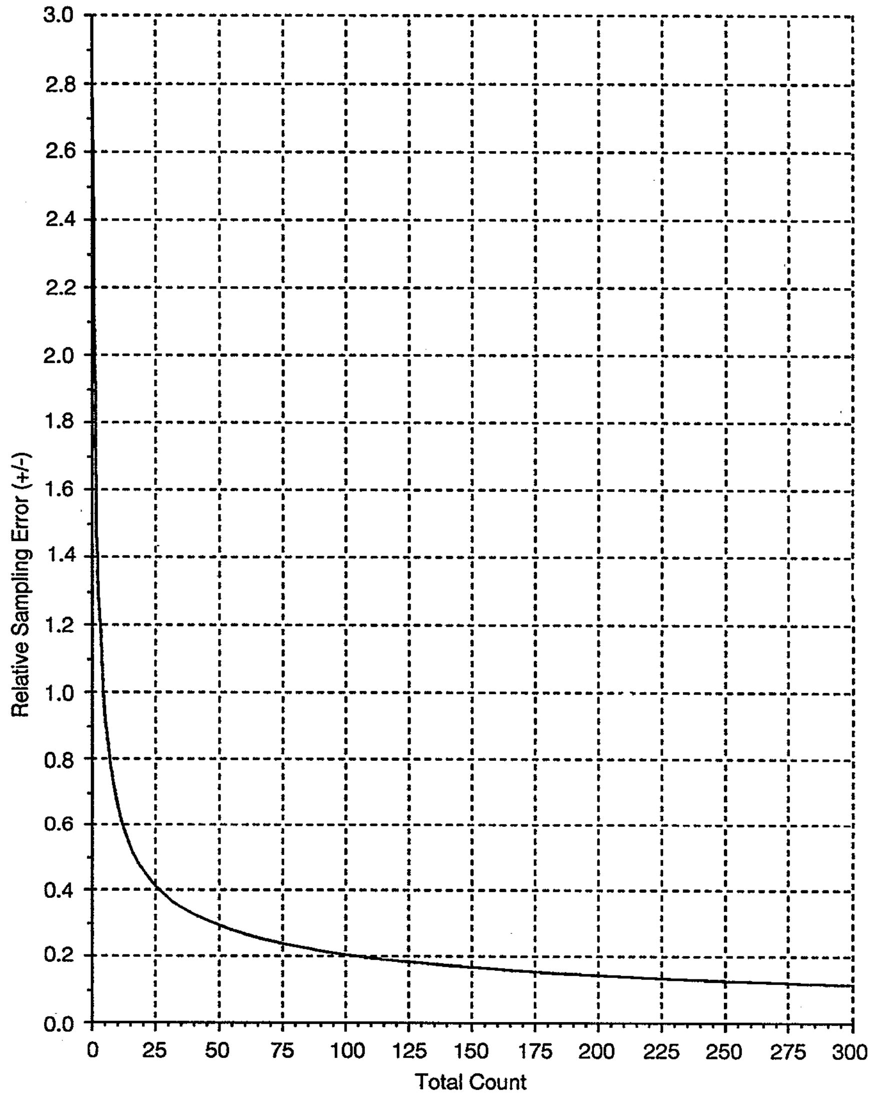


Figure 13-3. Relative sampling error of total plaque count half-length of 95% confidence interval divided by total plaque count, assuming randomly dispersed (Poisson) counts.

*U.S. Government Printing Office: 1993 — 750-002/80269