

Chapter 352

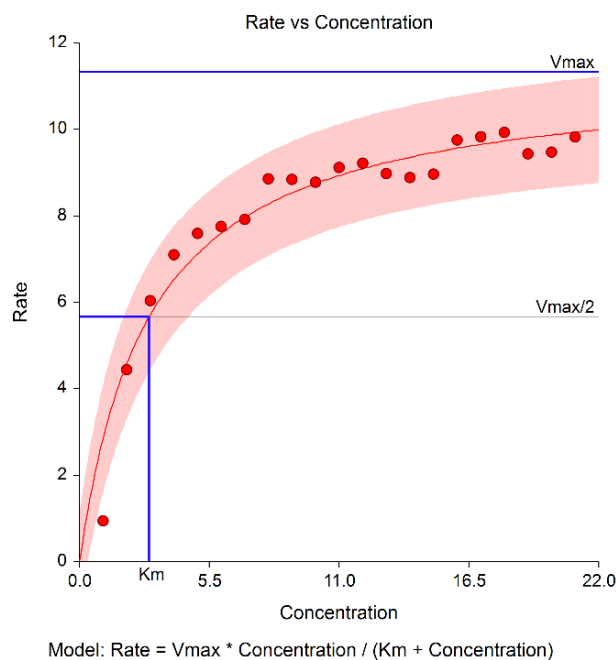
Michaelis-Menten Equation

Introduction

The Michaelis-Menten equation is a well-known model used in enzyme kinetics. It is a special arrangement of a two-parameter rectangular hyperbola. The mathematical model is

$$V = \frac{C(V_{max})}{C + K_m}$$

where V is the dependent variable, C is the independent variable, and V_{max} and K_m are parameters to be estimated. In enzyme kinetics, V is the velocity (rate) of an enzyme reaction and C is the substrate concentration. V_{max} and K_m have simple physical interpretations. V_{max} is the maximum velocity and serves as a horizontal asymptote. K_m , the Michaelis constant or ED50, is the value of C the results a velocity of $V_{max}/2$.



This provides new technologies for fitting and testing the parameters of the Michaelis-Menten equation that have not been easily available. First, it can fit several batches of data simultaneously. Second, it compares fitted models across batches using both graphics and numerical tests such as an approximate F-test for curve coincidence and a computer-intensive randomization test that compares curve coincidence and individual parameter values. Third, it fits both a maximum-likelihood and a nonlinear regression model. Fourth, it computes bootstrap confidence intervals for parameter values, predicted means, and predicted values using the latest computer-intensive bootstrapping technology.

Technical Details

Two methods of estimation are provided: nonlinear least-squares regression and maximum likelihood. These will be discussed next.

Nonlinear Regression

Estimation

Nonlinear regression is the algorithm used in **NCSS** to fit various nonlinear model. The nonlinear regression model associated with the Michaelis-Menten equation is

$$V = \frac{C(Vmax)}{C + Km} + \varepsilon$$

where ε represents normally distributed errors with zero mean and constant variance σ^2 . It provides estimates, confidence intervals, and statistical hypothesis tests based on this assumption. The method is documented in the chapter entitled *Introduction to Curve Fitting*. We refer you to that chapter for details.

Confidence Intervals

Two methods are used to calculate confidence intervals of the regression parameters and predicted values. The first method is based on the usual normality and constant variance of residuals assumption. When the data follow these assumptions, standard expressions for the confidence intervals are used based on the Student's t distribution. Unfortunately, nonlinear regression dataset rarely follow these assumptions.

The second method is called the *bootstrap* method. This is a modern, computer-intensive method that has only become available in recent years as extensive computer power has become available.

Bootstrap Confidence Intervals

Bootstrapping provides standard errors and confidence intervals for nonlinear-regression parameter, predicted means, and predicted values. The method is simple in concept, but it requires extensive computation time.

Bootstrap confidence intervals are based on the assumption that your sample is actually representative of the population. Beginning with this assumption, B samples are drawn (B is over 1000) of size N from your original sample with replacement. With replacement sampling means that each observation may be selected more than once. For each bootstrap sample, the nonlinear-regression results are computed and stored.

Suppose you want the standard error and a confidence interval of a regression parameter. The bootstrap sampling process provides B estimates of this parameter. The standard deviation of these B estimates is the bootstrap estimate of the standard error of the parameter. The bootstrap confidence interval is found by arranging the B values in sorted order and selecting the appropriate percentiles from the list. For example, a 90% bootstrap confidence interval for the parameter is given by fifth and ninety-fifth percentiles of the bootstrap parameter values.

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The main assumption made when using the bootstrap is that your sample approximates the population. Because of this assumption, bootstrapping does not work well for small samples in which there is little likelihood that the sample is representative of the population. Bootstrapping should only be used in medium to large samples.

Bootstrap Prediction Intervals

Bootstrap confidence intervals for the mean of Y given X are generated from the bootstrap sample in the usual way. To calculate prediction intervals for the predicted value (not the mean) of Y given X requires a modification to the predicted value of Y to be made to account for the variation of Y about its mean. This modification of the predicted Y values in the bootstrap sample, suggested by Davison and Hinkley, is as follows.

$$\hat{y}_i = \hat{y}_i + e_r^*$$

where e_r^* is a randomly selected modified residual (see below). By adding the residual, we have added an appropriate amount of variation to represent the variance of individual Y 's about their mean value.

Modified Residuals

Davison and Hinkley (1999) page 279 recommend the use of a special rescaling of the residuals when bootstrapping to keep results unbiased. Because of the high amount of computing involved in bootstrapping, these modified residuals are calculated using

$$e_j^* = \frac{e_j}{\sqrt{1 - \frac{1}{N}}} - \bar{e}$$

where

$$\bar{e} = \frac{\sum_{j=1}^N e_j}{N}$$

Note that there is a different rescaling than Davison and Hinkley recommended. We have used this rescaling because it is much quicker to calculate.

Hypothesis Testing

When curves are fit to two or more groups, it is often of interest to test whether certain regression parameters are equal and whether the fitted curves coincide. Although some approximate results have been obtained using indicator variables, these are asymptotic results, and little is known about their appropriateness in sample samples. We provide a test of the hypothesis that all group curves coincide using an F -test that compares the residual sum of squares obtained when the grouping is ignored with the total of the residual sum of squares obtained for each group. This test is routinely used in the analysis of variance associated with linear models and its application to nonlinear models has occasionally been suggested. However, it is based on naive assumptions that seldom occur.

Because of the availability of fast computing speed in recent years, a second method of hypothesis testing, called the *randomization test*, is now available. This test will be discussed next.

Randomization Test

Randomization testing is discussed by Edgington (1987). The details of the randomization test are simple: all possible permutations of the group variable while leaving the dependent and independent variables in their original order are investigated. For each permutation, the difference between the estimated group parameters is calculated. The number of permutations with a magnitude greater than or equal to that of the actual sample is counted. Dividing this count by the number of permutations gives the significance level of the test.

The randomization test is suggested because an exact test is achieved without making unrealistic assumptions about the data such as constant variance, normality, or model accuracy. The test was not used in the past because the number of computations was prohibitive. In fact, the randomization test was originally proposed by Fisher, and he chose his F -test because its distribution close approximated the randomization distribution.

The only assumption that a randomization test makes is that the data values are *exchangeable* under the null hypothesis.

For even moderate sample sizes, the total number of permutations is in the trillions, so a Monte Carlo approach is used in which the permutations are found by random selection rather than enumeration. Using this approach, a reasonable approximation to the test's probability level may be found by considering only a few thousand permutations rather than the trillions needed for complete enumeration. Edgington suggests that at least 1000 permutations be computed. We suggest that this be increased to 10000 for important results.

The program tests two types of hypotheses using randomization tests. The first is that each of the estimated model parameters is equal. The second is that the individual fitted curves coincide across all groups.

Randomization Statistics for Testing Parameter Equivalence

The test statistic for comparing a model parameter is formed by summing the difference between the group parameter estimates for each pair of groups. If there are G groups, the test statistic is computed using the formula

$$B_{RT} = \sum_{i=1}^{G-1} \sum_{j=i+1}^G |\hat{\beta}_i - \hat{\beta}_j|$$

Randomization Statistics for Testing Curve Equivalence

The test statistic for comparing the whole curve is formed by summing the difference between the estimated predicted values for each pair of groups at several points along the curve. If there are G groups and K equally spaced test points, the test statistic is computed using the formula

$$C_{RT} = \sum_{k=1}^K \sum_{i=1}^{G-1} \sum_{j=i+1}^G |\hat{y}_{ki} - \hat{y}_{kj}|$$

Maximum Likelihood

The maximum-likelihood estimation formulas are found in Raaijmakers (1987). This method leads to simple, analytical formulae for the parameters as well as large-sample confidence intervals.

The assumption of a constant absolute error made by nonlinear regression is often not appropriate. It is often more appropriate to assume that σ is proportional to the mean value at each value of C . This leads to the following statistical model

$$V = \frac{C(Vmax)}{C + Km} + \left(\frac{C(Vmax)}{C + Km} \right) \varepsilon$$

If we define X to be V/C , Raaijmakers (1987) provided the following estimates of the Michaelis-Menten parameters.

$$\widehat{Vmax} = \bar{V} + \widehat{Km} \bar{X}$$

$$\widehat{Km} = \frac{\bar{X}S_{VV} - \bar{V}S_{XV}}{\bar{V}S_{XX} - \bar{X}S_{XV}}$$

where S_{VV} , S_{XV} , and S_{XX} are the sum of squares and cross products of the deviations $V - \bar{V}$ and $X - \bar{X}$. \bar{V} and \bar{X} are the sample means of the corresponding variables.

An unbiased estimate of the error variance is given by

$$\hat{\sigma}^2 = \frac{S_{VV} + 2\widehat{Km}S_{XV} + \widehat{Km}^2S_{XX}}{N - 2}$$

where N is the sample size.

The large-sample variances of these estimates are given by

$$\text{var}(\widehat{Km}) \approx \frac{\sigma^2}{(1 + 2\sigma^2/\widehat{Vmax}^2) \sum_{i=1}^N (U_i - \bar{U})^2}$$

$$\text{var}(\widehat{Vmax}) \approx \frac{\sigma^2}{N} + \bar{U}^2 \text{var}(\widehat{Km})$$

where

$$U_i = \frac{Vmax}{C_i + Km}$$

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Large sample confidence intervals based on maximum-likelihood estimation can be calculated for these estimates. The $100(1 - \alpha)\%$ confidence interval for Km is

$$\widehat{Km} \pm z_{1-\alpha/2} \sqrt{\text{var}(\widehat{Km})}$$

The $100(1 - \alpha)\%$ confidence interval for $Vmax$ is

$$\widehat{Vmax} \pm z_{1-\alpha/2} \sqrt{\text{var}(\widehat{Vmax})}$$

The widths of these intervals are easily determined.

A close inspection of these formulas will show that they depend on the experimental design. That is, the C values. So, the sample size formulae require the specification of a specific design.

Data Structure

The data are entered in two variables: one dependent variable and one independent variable. Additionally, you may specify a frequency variable containing the observation count for each row and a group variable that is used to partition the data into independent groups.

Missing Values

Rows with missing values in the variables being analyzed are ignored in the calculations. When only the value of the dependent variable is missing, predicted values are generated.

The maximum-likelihood estimation procedure cannot tolerate concentrations of exactly zero since these values are used in the denominator. Hence, if such values occur, they are treated as missing values by the maximum-likelihood estimation. They are included during the nonlinear regression estimate.

Example 1 – Fitting the Michaelis-Menten Equation

This section presents an example of how to fit and compare a Michaelis-Menten model to two groups of data. This example will use the data in the MichaelisMenten dataset. In this example, the dependent variable is *Rate* and the independent variable is *Concentration*. The groups are defined by the values of *Type*.

Setup

To run this example, complete the following steps:

1 Open the MichaelisMenten example dataset

- From the File menu of the NCSS Data window, select **Open Example Data**.
- Select **MichaelisMenten** and click **OK**.

2 Specify the Michaelis-Menten Equation procedure options

- Find and open the **Michaelis-Menten Equation** procedure using the menus or the Procedure Navigator.
- The settings for this example are listed below and are stored in the **Example 1** settings file. To load these settings to the procedure window, click **Open Example Settings File** in the Help Center or File menu.

Variables Tab

Y (Rate) Variable **Rate**
 X (Concentration) Variable **Concentration**
 Group Variable..... **Type**
 Randomization Hypothesis Tests **Checked**

Reports Tab

Combined Summary Report **Checked**
 Group Difference F-Test **Checked**
 Iteration Summary Report..... **Checked**
 Iteration Detail Report..... **Unchecked**
 Maximum-Likelihood Estimation **Checked**
 Report
 Nonlinear-Regression Estimation **Checked**
 Report
 Analysis of Variance Report **Checked**
 Parameter Correlations Report..... **Checked**
 Predicted Values Report..... **Checked**
 Residual Report..... **Checked**
 Predict Y at these X Values **5 10 15 20**

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Plots Tab

Combined Function Plot Y**Checked**
 Individual Function Plots Y**Checked**
 Residual Plot Y**Checked**
 Probability Plot Y**Checked**
 Combined Function Plot Trans(Y).....**Unchecked**
 Individual Function Plot Trans(Y).....**Unchecked**
 Residual Plot Trans(Y).....**Unchecked**
 Probability Plot Trans(Y).....**Unchecked**

Resampling Tab

Samples (N).....**3000**
 Monte Carlo Samples**1000**
 Random Seed.....**11323** (for reproducibility)

3 Run the procedure

- Click the **Run** button to perform the calculations and generate the output.

Parameter Estimates for All Groups**Parameter Estimates for All Groups**

Type	Count	Iter's	R ²	Nonlinear Regression Vmax	Maximum Likelihood Vmax	Nonlinear Regression Km	Maximum Likelihood Km
1	21	6	0.97819	12.15445	13.13860	8.02219	10.05605
3	21	9	0.93628	11.33055	11.60003	2.95778	3.49882
Combined	42	8	0.80187	11.33658	10.98392	4.57479	4.49856

This report displays a summary of the results for each group and then for the case in which all groups are combined into one group.

Type

This column, headed by the name of the Group Variable, lists the group value that is displayed on this line. Note that the Value Labels option may be used to give more meaningful names to these values.

Count

This is the number of observations used by the nonlinear regression algorithm.

Iter's

This is the number of iterations used by the nonlinear regression algorithm to find the estimates. You should note whether the maximum number of iterations has been reached (in which case the algorithm did not converge).

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R²

This is the value of the pseudo R-squared value. A value near one indicates that the model fits the data well. A value near zero indicates that the model does not fit the data well.

Nonlinear Regression Vmax and Km

The nonlinear regression estimates of Vmax and Km are provided.

Maximum Likelihood Vmax and Km

The maximum likelihood estimates of Vmax and Km are provided.

Analysis of Variance Across Groups
Analysis of Variance Across Groups

Type	Count	Iter's	Model R ²	Error DF	Sum Squares Error	Mean Square Error
1	21	6	0.97819	19	2.29790	0.12094
3	21	9	0.93628	19	5.85704	0.30827
Combined	42	8	0.80187	40	45.98386	1.14960
Ignored			0.96486	38	8.15493	0.21460

This report displays goodness of fit results for each group and then for the case in which all groups are combined into one dataset. The final row of the report, labeled 'Ignored', gives the goodness of fit statistics for the model in which a separate curve is fit for each group.

Group Name (Type)

This column, headed by the name of the Group Variable, lists the group value that is displayed on this line.

Count

This is the number of observations used by the nonlinear regression algorithm.

Iter's

This is the number of iterations used by the nonlinear regression algorithm to find the estimates. You should note whether the maximum number of iterations has been reached (in which case the algorithm did not converge).

R²

This is the value of the pseudo R-squared value. A value near one indicates that the model fits the data well. A value near zero indicates that the model does not fit the data well. Note

Error DF

The degrees of freedom are the number of observations minus the number of parameters fit.

Sum Squares Error

This is the sum of the squared residuals for this group.

Mean Square Error

This is a rough estimate of the variance of the residuals for this group.

Curve Inequality F-Test

Curve Inequality F-Test

Curves Tested	DF	Mean Square	F Ratio	F-Test Prob Level
All	2	18.91446	88.1368	0.00000
Error	38	0.21460		

This report displays an F-Test of whether all of the group curves are equal. This test compares the residual sum of squares obtained when the grouping is ignored with the total of the residual sum of squares obtained for each group. This test is routinely used in analysis linear models and its application to nonlinear models has occasionally been suggested. However, it is based on normality assumptions which seldom occur. When testing curve coincidence is important, we suggest you use a randomization test.

Curves Tested

This column indicates the term presented on this row.

DF

The degrees of freedom of this term.

Mean Square

The mean square associated with this term.

F Ratio

The F-ratio for testing the hypothesis that all curves coincide.

F-Test Prob Level

This is the probability level of the F-ratio. When this value is less than 0.05 (a common value for alpha), the test is 'significant' meaning that the hypothesis of equal curves is rejected. If this value is larger than the nominal level (0.05), the null hypothesis cannot be rejected. We do not have enough evidence to reject.

Curve Inequality Randomization Tests

Curve Inequality Randomization Tests

Curves Tested	Randomization Prob Level	Monte Carlo Samples	Number of Points Compared Along Curve
1 vs. 3	0.00000	1000	10

User-Entered Random Seed: 11323

This report displays the results of a randomization test whose null hypothesis is that the all the group curves coincide. When more than two groups are present, a separate test is provided for each pair of groups, plus a combined test of the equality of all groups.

Curves Tested

This column indicates the groups whose equality is being test on this row.

Randomization Prob Level

This is the two-sided probability level of the randomization test. When this value is less than 0.05, the test is 'significant' meaning that the null hypothesis of equal curves is rejected. If this value is larger than the nominal level (0.05), there is not enough evidence in the data to reject the null hypothesis of equality.

(Note: because this is a Monte Carlo test, your results may vary from those displayed here.)

Monte Carlo Samples

The number of Monte Carlo samples.

Number of Points Compared Along the Curve

The number of values along the X axis at which a comparison between curves is made. Of course, the more X values used, the more accurate (and time consuming) will be the test.

Parameter Inequality Randomization Tests

Parameter Inequality Randomization Tests

Curves Compared	Parameter Tested	Randomization Prob Level	Monte Carlo Iterations
1 vs. 3	Vmax	0.52200	1000
1 vs. 3	Km	0.01800	1000

User-Entered Random Seed: 11323

This report displays the results of randomization tests about the equality of each parameter across groups. When more than two groups are present, a separate test is provided for each pair of groups, plus a combined test of parameter equality of all groups.

Curves Compared

This column indicates the groups being test on this row.

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Parameter Test

This column indicates model parameter whose equality is being tested.

Randomization Prob Level

This is the two-sided probability level of the randomization test. When this value is less than 0.05, the test is 'significant' meaning that the null hypothesis of equal parameter values across groups is rejected. If this value is larger than the nominal level (0.05), there is not enough evidence in the data to reject the null hypothesis of equality.

(Note: because this is a Monte Carlo test, your results may vary from those displayed here.)

Monte Carlo Samples

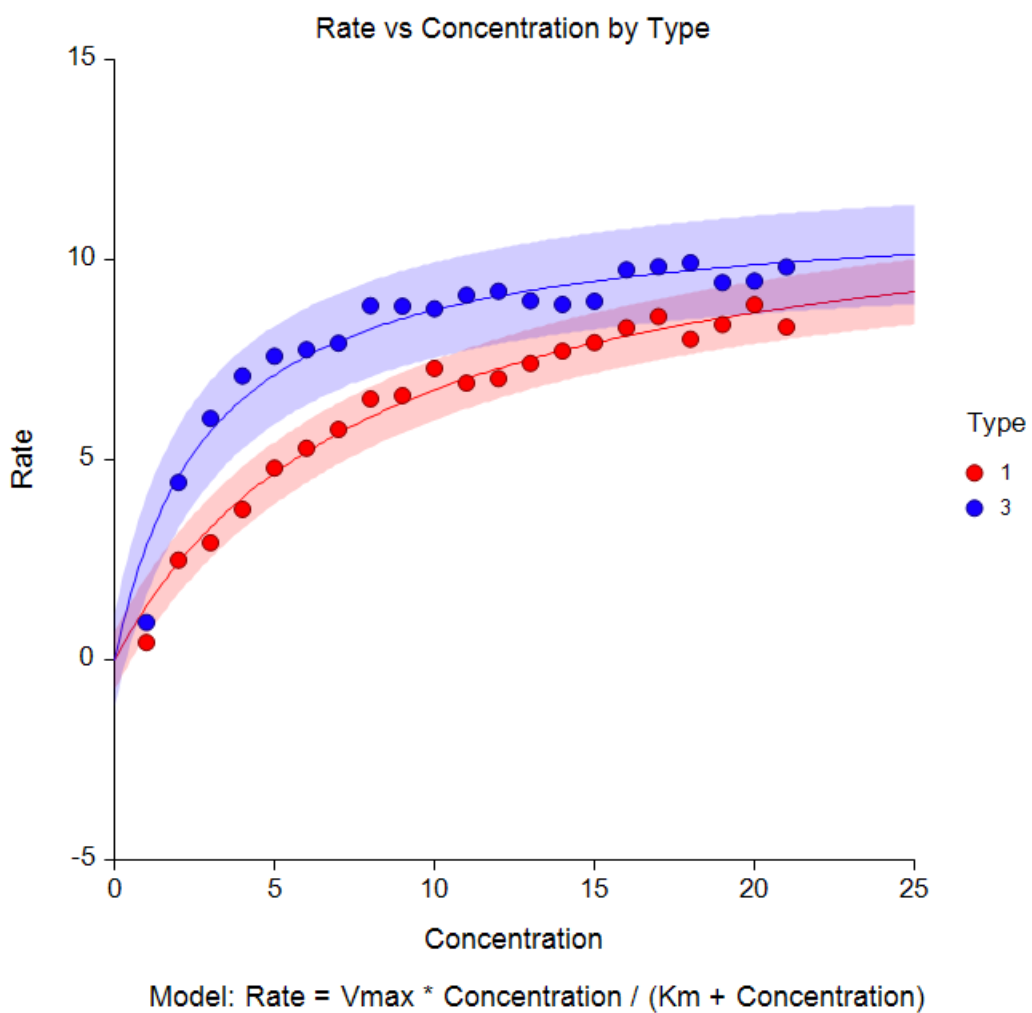
The number of Monte Carlo samples.

Number of Points Compared Along the Curve

The number of values along the X axis at which a comparison between curves is made. Of course, the more X values used, the more accurate (and time consuming) will be the test.

Combined Function Plot

Combined Function Plot



This plot displays all of the data and fitted curves, allowing you to quickly assess the quality of the results.

Iteration Summary Section for Type=1

Iteration Summary Section for Type = 1

Iteration Number	Residual Sum of Squares	Vmax	Km
1	2.36176	12.15872	8.19424
2	2.29797	12.14473	8.00369
3	2.29790	12.15492	8.02304
4	2.29790	12.15445	8.02219
5	2.29790	12.15445	8.02219
6	2.29790	12.15445	8.02219

This report displays the progress of the search algorithm in its search for a solution. It allows you to assess whether the algorithm had indeed converged or whether the program should be re-run with the Maximum Iterations increased or the model changed.

Note that if over ten iterations were needed, the program does not display every iteration.

Maximum-Likelihood Parameter Estimation Section for Type = 1

Maximum-Likelihood Parameter Estimation Section for Type = 1

Assuming the Error Variance is Proportional to the Mean

Parameter Name	Parameter Estimate	Asymptotic Standard Error	Lower 95% C.L.	Upper 95% C.L.
Vmax	13.13860	1.45928	10.08430	16.19290
Km	10.05605	2.02340	5.82102	14.29107

Data Summary

Total Count	21
Total Count Used	21

Note: Rows ignored if X = 0.

This report displays the details of the maximum-likelihood estimation of the model parameters.

Parameter Name

The name of the parameter whose results are shown on this line.

Parameter Estimate

The estimated value of this parameter.

Asymptotic Standard Error

An estimate of the standard error from maximum-likelihood results.

Lower 95% C.L.

The lower value of a 95% confidence limit for this parameter.

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Upper 95% C.L.

The upper value of a 95% confidence limit for this parameter.

Nonlinear Regression Parameter Estimation Section for Type = 1

Nonlinear Regression Parameter Estimation Section for Type = 1

Assuming Constant Error Variance

Parameter Name	Parameter Estimate	Asymptotic Standard Error	Lower 95% C.L.	Upper 95% C.L.
Vmax	12.15445	0.50850	11.09015	13.21875
Km	8.02219	0.83916	6.26580	9.77858

Model Estimation Information

Iterations	6	Rows Read	21
R ²	0.978188	Rows Used	21
Random Seed	11323	Total Count	21

Estimated Nonlinear-Regression Model

$(12.1544486443541) * (\text{Concentration}) / ((8.02219181570417) + (\text{Concentration}))$

This report displays the details of the estimation of the model parameters.

Parameter Name

The name of the parameter whose results are shown on this line.

Parameter Estimate

The estimated value of this parameter.

Asymptotic Standard Error

An estimate of the standard error of the parameter based on asymptotic (large sample) results.

Lower 95% C.L.

The lower value of a 95% confidence limit for this parameter. This is a large sample (at least 25 observations for each parameter) confidence limit. In most cases, the bootstrap confidence interval will be more accurate.

Upper 95% C.L.

The upper value of a 95% confidence limit for this parameter. This is a large sample (at least 25 observations for each parameter) confidence limit. In most cases, the bootstrap confidence interval will be more accurate.

Iterations

The number of iterations that were completed before the nonlinear algorithm terminated. If the number of iterations is equal to the Maximum Iterations that you set, the algorithm did not converge, but was aborted.

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R²

There is no direct R-squared defined for nonlinear regression. This is a pseudo R-squared constructed to approximate the usual R-squared value used in multiple regression. We use the following generalization of the usual R-squared formula:

$$R\text{-Squared} = (ModelSS - MeanSS)/(TotalSS - MeanSS)$$

where *MeanSS* is the sum of squares due to the mean, *ModelSS* is the sum of squares due to the model, and *TotalSS* is the total (uncorrected) sum of squares of Y (the dependent variable).

This version of R-squared tells you how well the model performs after removing the influence of the mean of Y. Since many nonlinear models do not explicitly include a parameter for the mean of Y, this R-squared may be negative (in which case we set it to zero) or difficult to interpret. However, if you think of it as a direct extension of the R-squared that you use in multiple regression, it will serve well for comparative purposes.

Random Seed

This is the value of the random seed that was used when running the bootstrap confidence intervals and randomization tests. If you want to duplicate your results exactly, enter this random seed into the Random Seed box under the Simulation tab.

Estimated Model

This is the model that was estimated with the parameters replaced with their estimated values. This expression may be copied and pasted as a variable transformation in the spreadsheet. This will allow you to predict for additional values of X. Note that to ensure accuracy, the parameter estimates are always given to double-precision accuracy.

Analysis of Variance Table for Type = 1
Analysis of Variance Table for Type = 1

Source	DF	Sum of Squares	Mean Square
Mean	1	847.88494	847.88494
Model	2	950.93664	475.46832
Model (Adjusted)	1	103.05170	103.05170
Error	19	2.29790	0.12094
Total (Adjusted)	20	105.34959	
Total	21	953.23453	

Source

The labels of the various sources of variation.

DF

The degrees of freedom.

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Sum of Squares

The sum of squares associated with this term. Note that these sums of squares are based on Y, the dependent variable. Individual terms are defined as follows:

Mean	The sum of squares associated with the mean of Y. This may or may not be a part of the model. It is presented since it is the amount used to adjust the other sums of squares.
Model	The sum of squares associated with the model.
Model (Adjusted)	The model sum of squares minus the mean sum of squares.
Error	The sum of the squared residuals. This is often called the sum of squares error or just "SSE."
Total	The sum of the squared Y values.
Total (Adjusted)	The sum of the squared Y values minus the mean sum of squares.

Mean Square

The sum of squares divided by the degrees of freedom. The Mean Square for Error is an estimate of the underlying variation in the data.

Bootstrap Section for Type = 1

Bootstrap Section when Type = 1

Estimation Results		Bootstrap Confidence Limits		
Parameter	Estimate	Conf. Level	Lower	Upper
Vmax				
Original Value	12.15445	0.90000	11.29151	12.83884
Bootstrap Mean	12.19446	0.95000	11.11946	12.96140
Bias (BM - OV)	0.04001	0.99000	10.80199	13.23536
Bias Corrected	12.11444			
Standard Error	0.47077			
Km				
Original Value	8.02219	0.90000	6.61419	9.16985
Bootstrap Mean	8.09265	0.95000	6.31216	9.38324
Bias (BM - OV)	0.07046	0.99000	5.76488	9.77080
Bias Corrected	7.95174			
Standard Error	0.77561			
Predicted Mean and Confidence Limits of Rate When Concentration = 5.00000				
Original Value	4.66682	0.90000	4.48908	4.85373
Bootstrap Mean	4.66297	0.95000	4.45215	4.88663
Bias (BM - OV)	-0.00385	0.99000	4.36885	4.95193
Bias Corrected	4.67067			
Standard Error	0.11063			

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Predicted Mean and Confidence Limits of Rate When Concentration = 10.00000

Original Value	6.74416		0.90000	6.63046	6.87422
Bootstrap Mean	6.74158		0.95000	6.61108	6.89587
Bias (BM - OV)	-0.00258		0.99000	6.56320	6.94613
Bias Corrected	6.74674				
Standard Error	0.07392				

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(Report continues for the other values of Concentration)

Sampling Method = Observation, Confidence Limit Type = Reflection, Number of Samples = 3000,
User-Entered Random Seed = 11323.

This report provides bootstrap estimates and confidence intervals for the parameters, predicted means, and predicted values. Note that bootstrap confidence intervals and prediction intervals are provided for each of the X (Temp) value requested. Details of the bootstrap method were presented earlier in this chapter.

Original Value

This is the parameter estimate obtained from the complete sample without bootstrapping.

Bootstrap Mean

This is the average of the parameter estimates of the bootstrap samples.

Bias (BM - OV)

This is an estimate of the bias in the original estimate. It is computed by subtracting the original value from the bootstrap mean.

Bias Corrected

This is an estimated of the parameter that has been corrected for its bias. The correction is made by subtracting the estimated bias from the original parameter estimate.

Standard Error

This is the bootstrap method's estimate of the standard error of the parameter estimate. It is simply the standard deviation of the parameter estimate computed from the bootstrap estimates.

Conf. Level

This is the confidence coefficient of the bootstrap confidence interval given to the right.

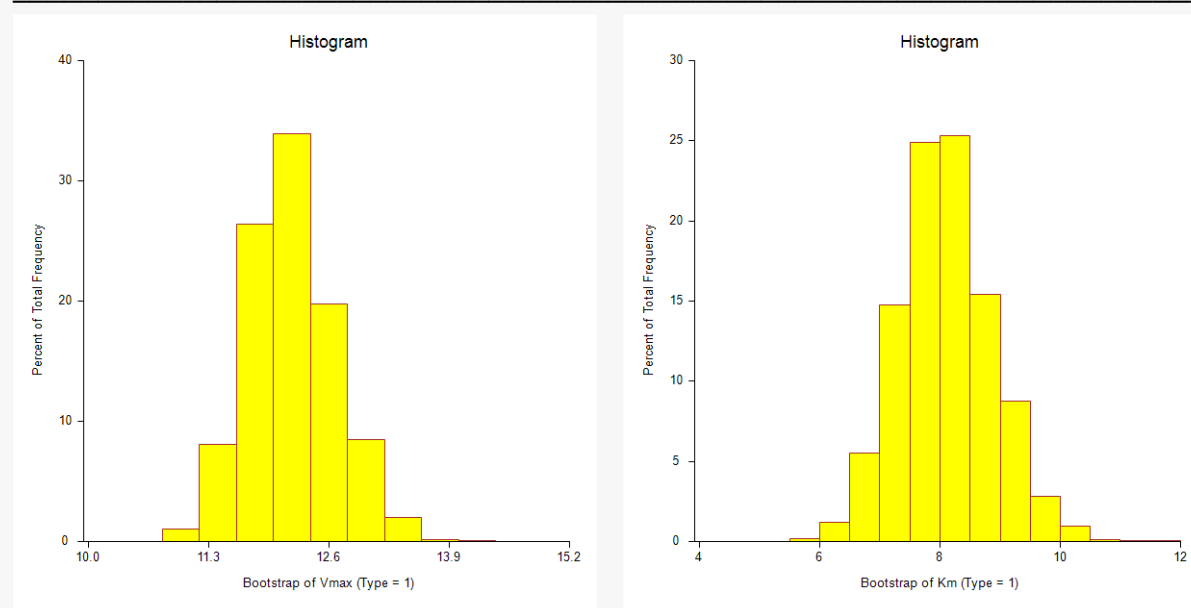
Bootstrap Confidence Limits - Lower and Upper

These are the limits of the bootstrap confidence interval with the confidence coefficient given to the left. These limits are computed using the confidence interval method (percentile or reflection) designated on the Bootstrap panel.

Note that to be accurate, these intervals must be based on over a thousand bootstrap samples and the original sample must be representative of the population.

Bootstrap Histograms for Type = 1

Bootstrap Histograms for Type = 1



(Several more histograms are displayed.)

Each histogram shows the distribution of the corresponding estimate.

Asymptotic Correlation Matrix of Parameters

Asymptotic Correlation Matrix of Parameters for Type = 1

	Vmax	Km
Vmax	1.000000	0.962983
Km	0.962983	1.000000

This report displays the asymptotic correlations of the parameter estimates. When these correlations are high (absolute value greater than 0.98), the precision of the parameter estimates is suspect.

Predicted Values for Specified X Values for Type=1

Predicted Values for Specified X Values for Type = 1

Concentration	Predicted Value of Rate	Lower 95.0% Prediction Limit	Upper 95.0% Prediction Limit
5.00000	4.66682	3.89418	5.43947
10.00000	6.74416	5.99394	7.49437
15.00000	7.91917	7.16492	8.67343
20.00000	8.67487	7.89564	9.45411

This section shows the predicted mean values and asymptotic (large sample) prediction intervals for the X values that were specified. Note that these are prediction limits for a new value, not confidence limits for the mean of the values.

Predicted Values and Residuals Section

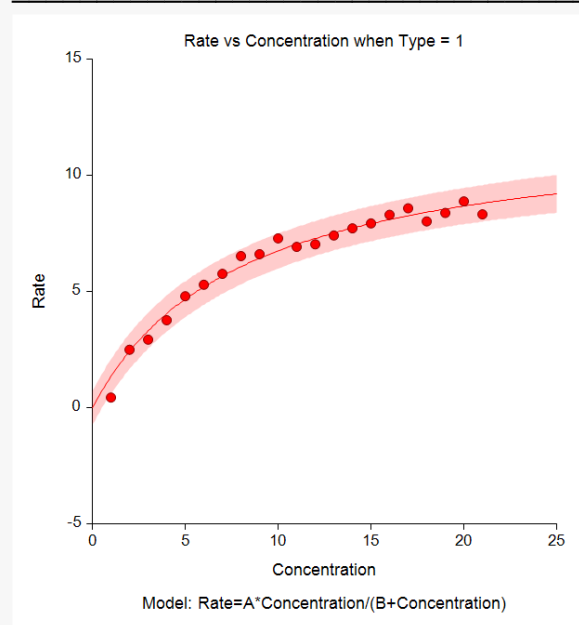
Predicted Values and Residuals Section for Type = 1

Row No.	Concentration	Rate	Predicted Value	Lower 95.0% Prediction Limit	Upper 95.0% Prediction Limit	Residual
1	1.00000	0.43846	1.34717	0.60374	2.09060	-0.90871
2	2.00000	2.49732	2.42551	1.66313	3.18788	0.07181
3	3.00000	2.93207	3.30818	2.53561	4.08075	-0.37610
4	4.00000	3.76707	4.04400	3.26899	4.81902	-0.27693
5	5.00000	4.79763	4.66682	3.89418	5.43947	0.13081
6	6.00000	5.29474	5.20081	4.43291	5.96870	0.09394
7	7.00000	5.76244	5.66370	4.90125	6.42614	0.09875
8	8.00000	6.52577	6.06881	5.31147	6.82614	0.45697
9	9.00000	6.60812	6.42632	5.67316	7.17948	0.18180
10	10.00000	7.28844	6.74416	5.99394	7.49437	0.54428
11	11.00000	6.92396	7.02858	6.27998	7.77718	-0.10461
12	12.00000	7.03491	7.28459	6.53629	8.03289	-0.24968
13	13.00000	7.41367	7.51624	6.76702	8.26546	-0.10257
14	14.00000	7.72145	7.72685	6.97560	8.47811	-0.00540
15	15.00000	7.93444	7.91917	7.16492	8.67343	0.01526
16	16.00000	8.30333	8.09548	7.33740	8.85356	0.20785
17	17.00000	8.58488	8.25769	7.49509	9.02030	0.32719
18	18.00000	8.01975	8.40744	7.63973	9.17516	-0.38769
19	19.00000	8.38369	8.54611	7.77282	9.31939	-0.16242
20	20.00000	8.88123	8.67487	7.89564	9.45411	0.20636
21	21.00000	8.32417	8.79477	8.00930	9.58023	-0.47059

This section shows the values of the predicted values, prediction limits, and residuals. If you have observations in which the independent variable is given, but the dependent (Y) variable is blank, a predicted value and prediction limits will be generated and displayed in this report.

Function Plot(s) for Type = 1

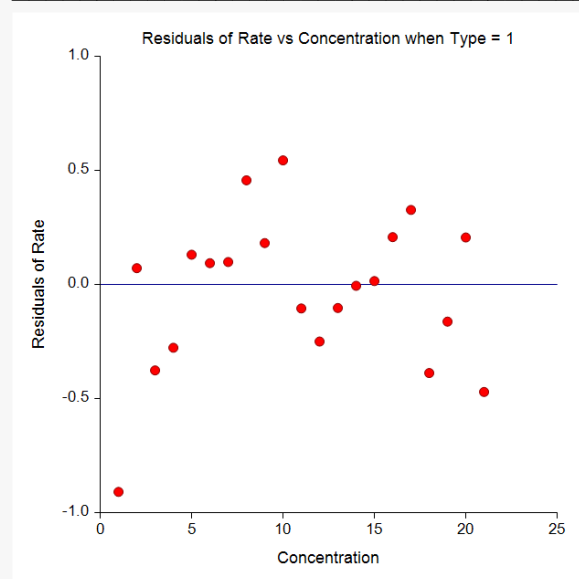
Function Plot(s) for Type = 1



This plot displays the data along with the estimated function. It is useful in deciding if the fit is adequate and the prediction limits are appropriate.

Residual Plot(s) for Type = 1

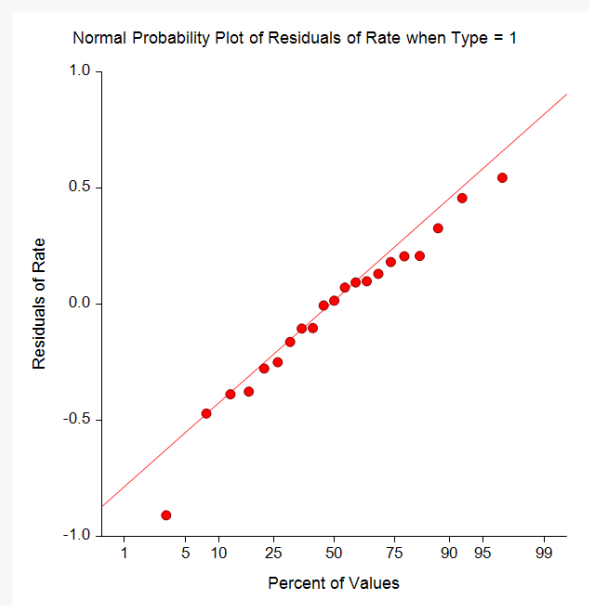
Residual Plot(s) for Type = 1



This is a scatter plot of the residuals versus the independent variable, X. The preferred pattern is a rectangular shape or point cloud. Any nonrandom pattern may require a redefinition of the model.

Probability Plot(s) for Type = 1

Probability Plot(s) for Type = 1



If the residuals are normally distributed, the data points of the normal probability plot will fall along a straight line. Major deviations from this ideal picture reflect departures from normality. Stragglers at either end of the normal probability plot indicate outliers, curvature at both ends of the plot indicates long or short distributional tails, convex or concave curvature indicates a lack of symmetry, and gaps or plateaus or segmentation in the normal probability plot may require a closer examination of the data or model. We do not recommend that you use this diagnostic with small sample sizes.