MEMLET Tutorial

Contents

Tutorial Introduction/Setup

Setting up the Program when Matlab is installed

1. Extract the supplied MEMLET Folder to a location of your choice
2. For the program to run, the current Matlab working directory must be the MEMLET folder, or the MEMLET folder must be added to the Matlab Path. It is recommended you add the folder to the Matlab path as described in steps 3 and 4 below.
3. Add the program files to the path by either:
   a. Changing the current Matlab working directory to the MEMLET folder then typing "addpath(pwd)" in the Matlab Command Window.

      OR

   b. In the command window, typing "addpath DIRNAME" where DIRNAME is replaced by the full path to the program directory (for example "addpath C:/Matlab Code/MEMLET/")

4. Type "savepath" into the Matlab Command window to ensure the MEMLET folder will be in the Matlab path each time Matlab opens.
5. Type MEMLET in the command window, or run MEMLET.m to run the program.

Setting up the Program when Matlab is *not* installed (Standalone)

1. Install the program by double clicking the Install MEMLET icon
2. Follow the installation instructions. If Matlab Runtime is not already installed, the program will prompt you for permission to automatically download and install it. **The Runtime from MathWorks is large and may take a while to download.**
3. Open the Program by navigating the installation path chosen during setup, and double clicking on MEMLET.exe inside of the "application" folder.

Note: Loading data using the "Enter Variable Name" function does not operate in a standalone installation. Also, the fitted values must be manually copied out of the program and are not saved in the workspace.

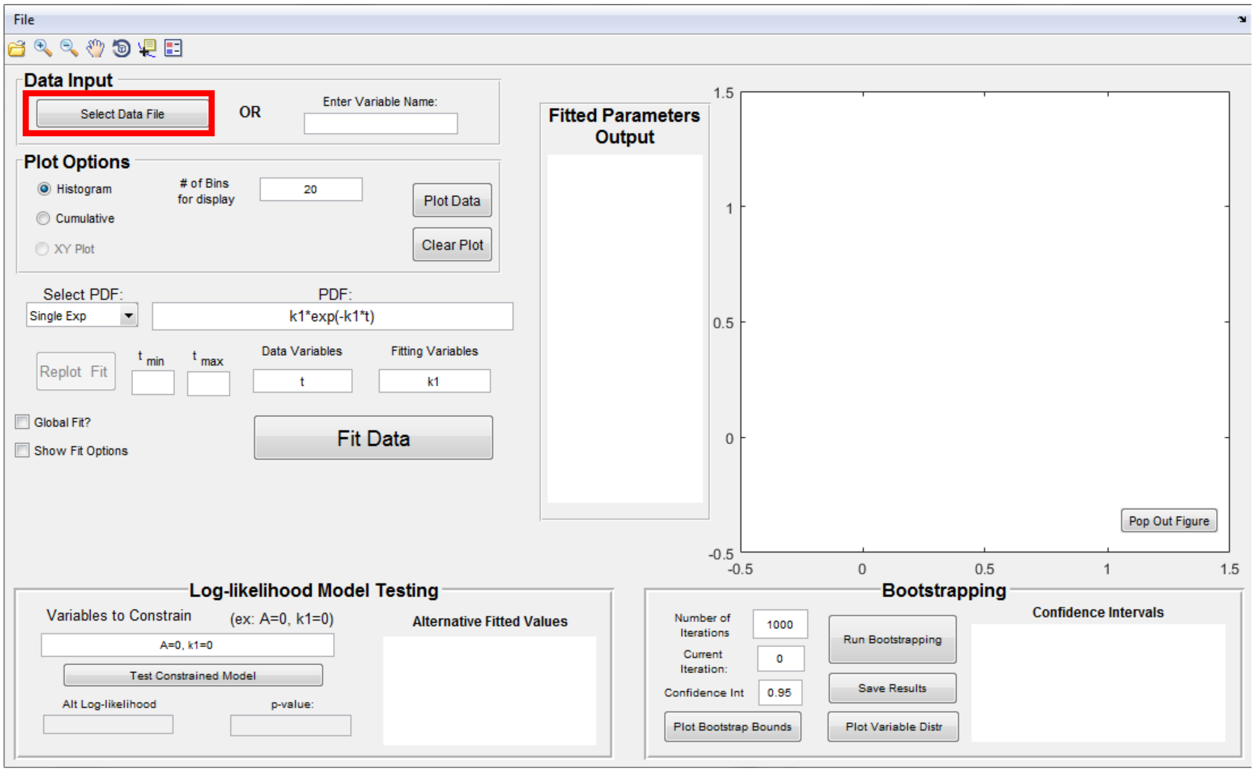
Sample Data files are located in the "Demo Data" Folder

Note on File Types:

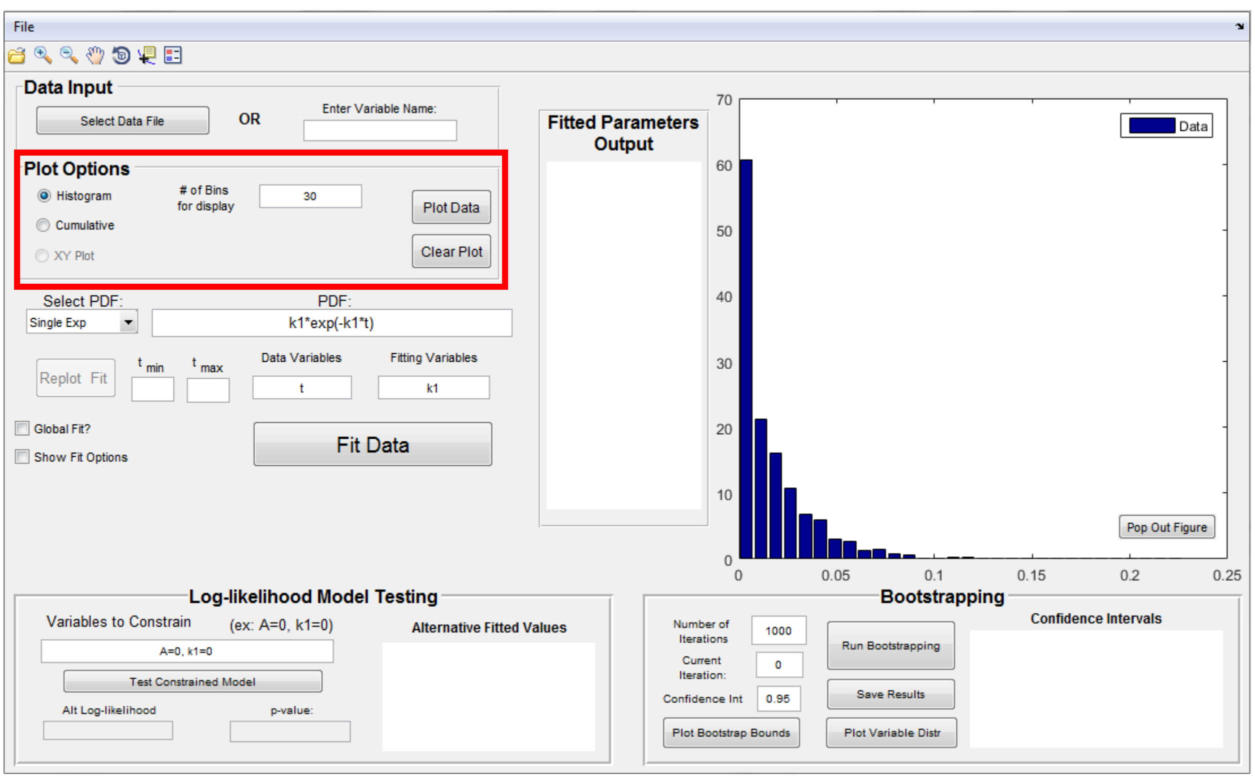
The program will read data from text files that contain lists of numbers in columns (i.e. each value on a new line). To load data with multiple dependent variables or datasets, arrange each set of values into its own column, with columns separated by either spaces, tabs, or commas. Saving an Excel file as a "CSV (comma separated)" file will produce a file that the program can read. Open the files in the Tutorial Datasets folder to see examples.

Tutorial 1: Performing a simple fit

1. Load Sample Double Exponential Data.
   a. Click *Select Data File*

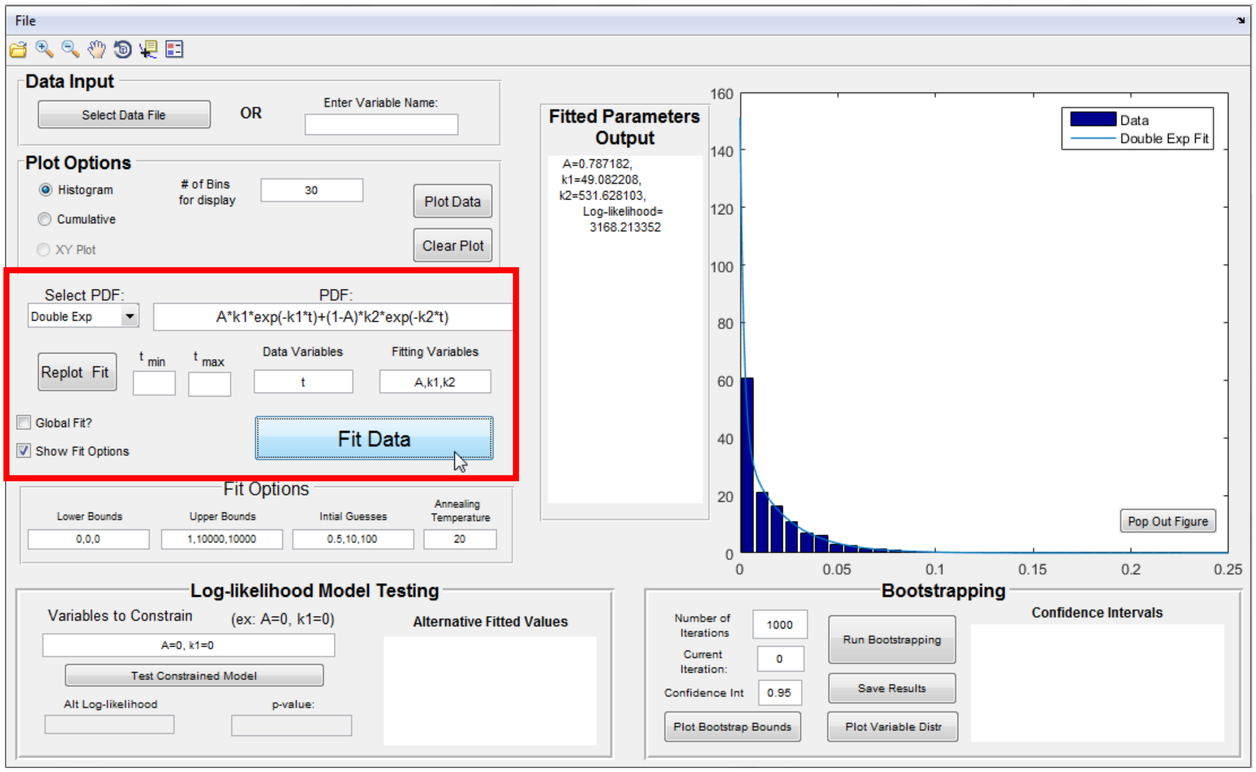


   b. Navigate to "Demo Data" Folder
   c. Open "Double Exponential Data.txt"
2. Plot Data
   a. Select *Histogram* under plot options (default)
   b. Choose 30 bins by typing 30 into the *# of Bins for Display* box
   c. Click *Plot Data*



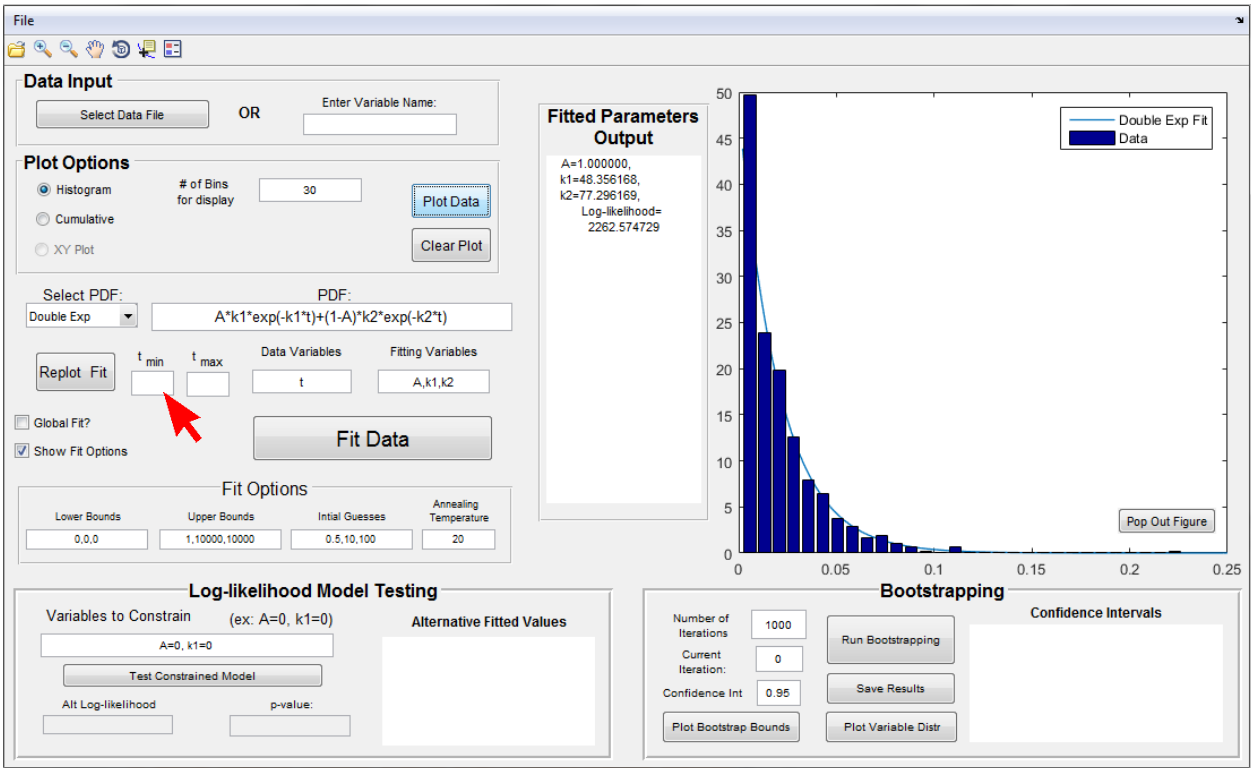
3. Fit Data

a. Choose "*Double Exp*" from the *Select PDF* drop down menu
b. Edit the Initial guesses and bounds, if desired, after clicking *Show Fit Options* (not required)
c. Click *Fit Data*
d. The fit will be plotted over the histogram and the parameter values will be given in the *Fitted Parameters Output* box

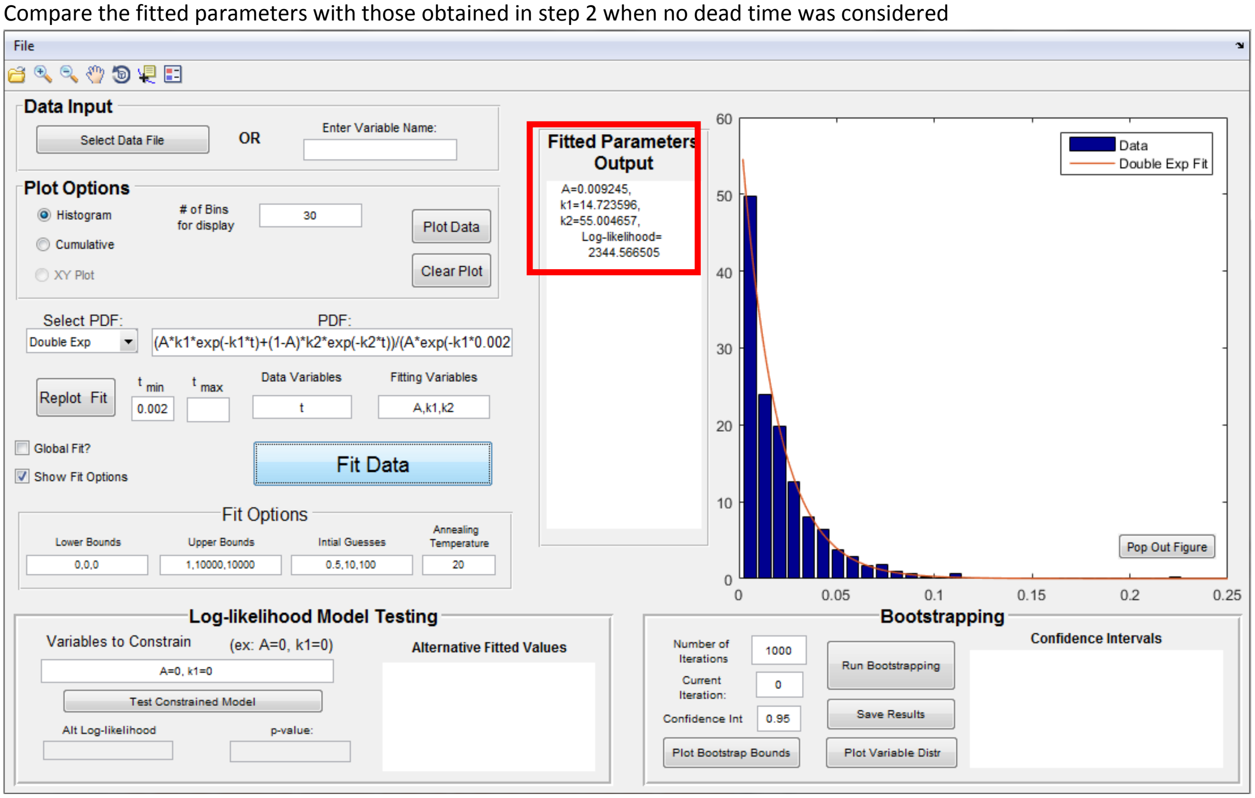


Tutorial 2: Performing a fit subject to a minimum detectable event

1. Load Sample Double Exponential Data with minimum detectable event by repeating steps 1-2 from tutorial 1, but selecting the "Double Exp with 2 ms DT.txt" sample data file.
2. Leave the "t_{min}" box blank and click *Fit Data*. Compare this new value for k2 to the value you obtained earlier when the data was not truncated by an imposed dead time.

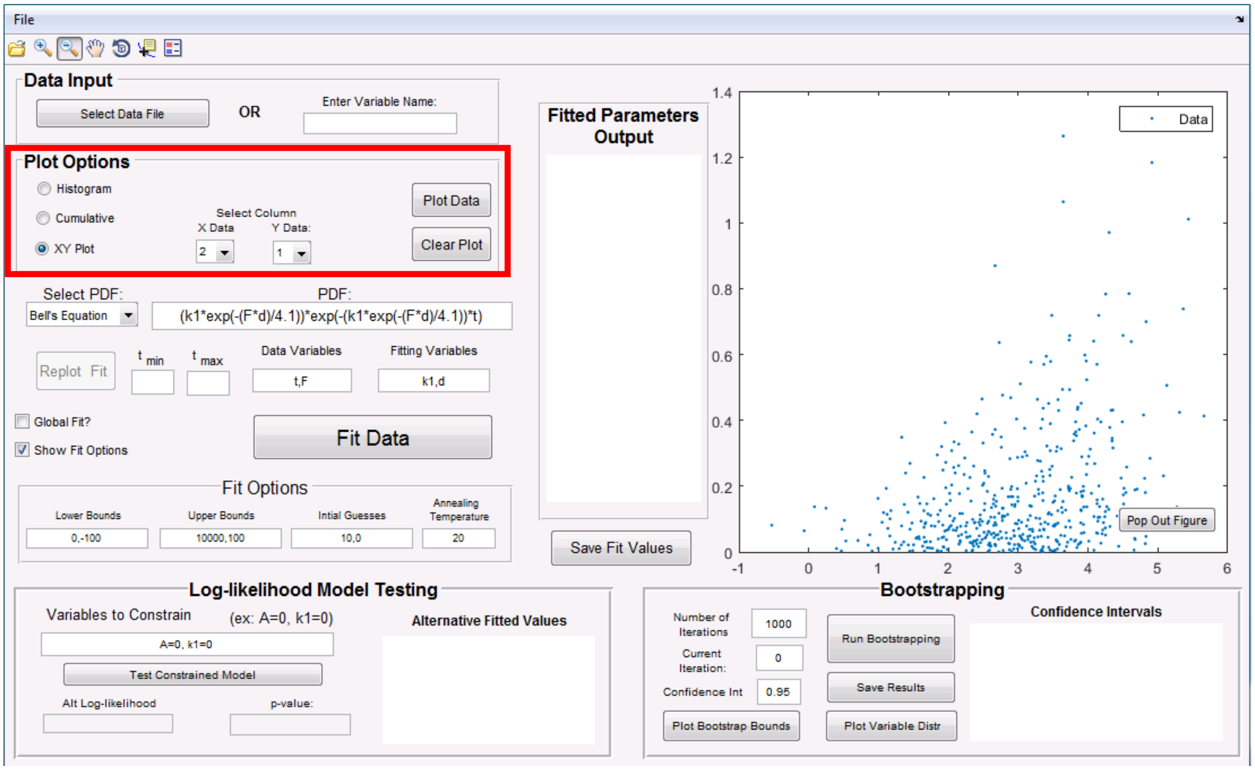


3. Enter "0.002" in the t_{min} box to account for the 2 ms dead time
4. Click *Fit Data*
5. Compare the fitted parameters with those obtained in step 2 when no dead time was considered

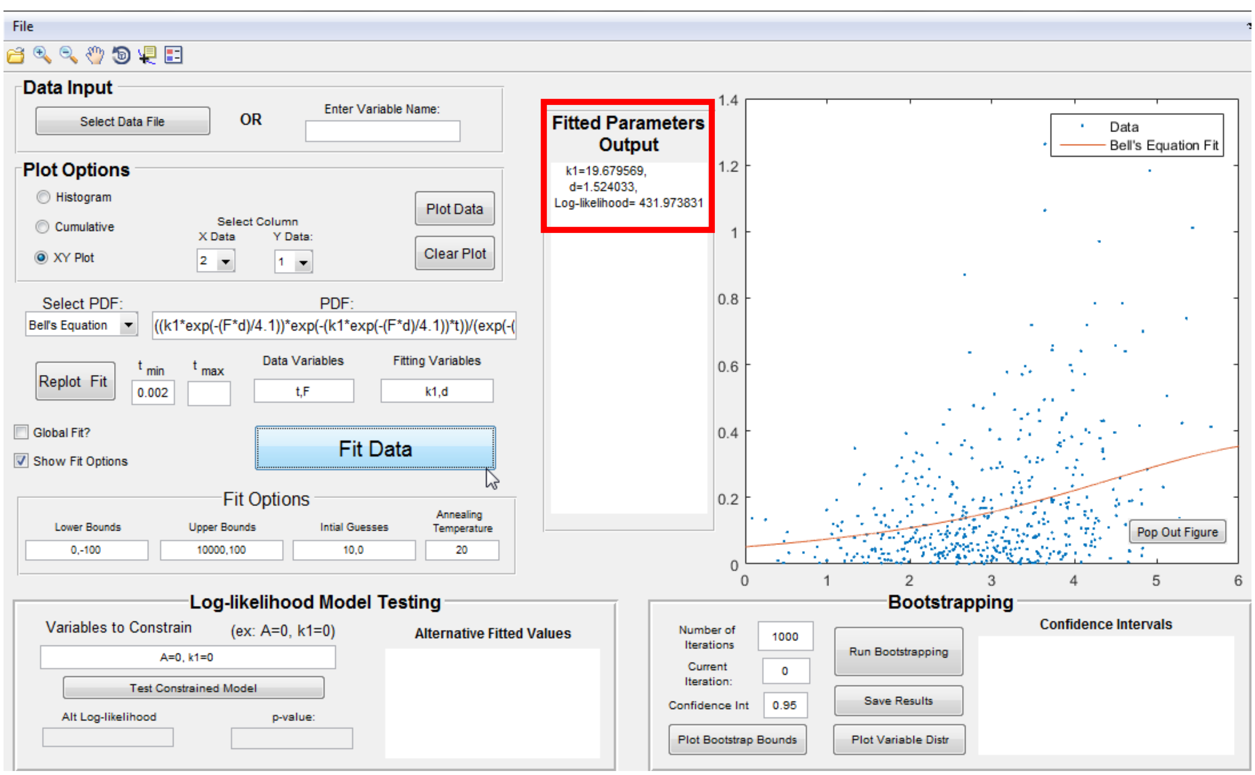


Tutorial 3: Fitting 2D datasets

1. Load the “2D Data Demo.txt” example data set. This contains a list of durations in the first column and list of corresponding forces in the second column.
2. Select *Bell’s Equation* from the *Select PDF* drop down menu and ensure t_{min} and t_{max} are blank (X-Y plots with a t_{min} or t_{max} require a 2D PDF to be selected to avoid an error).
   3. Select *X-Y plot* and choose *X Col* =2 and *Y Col*=1 to plot Forces on the x-axis and durations on the y-axis. Click *Plot Data*.

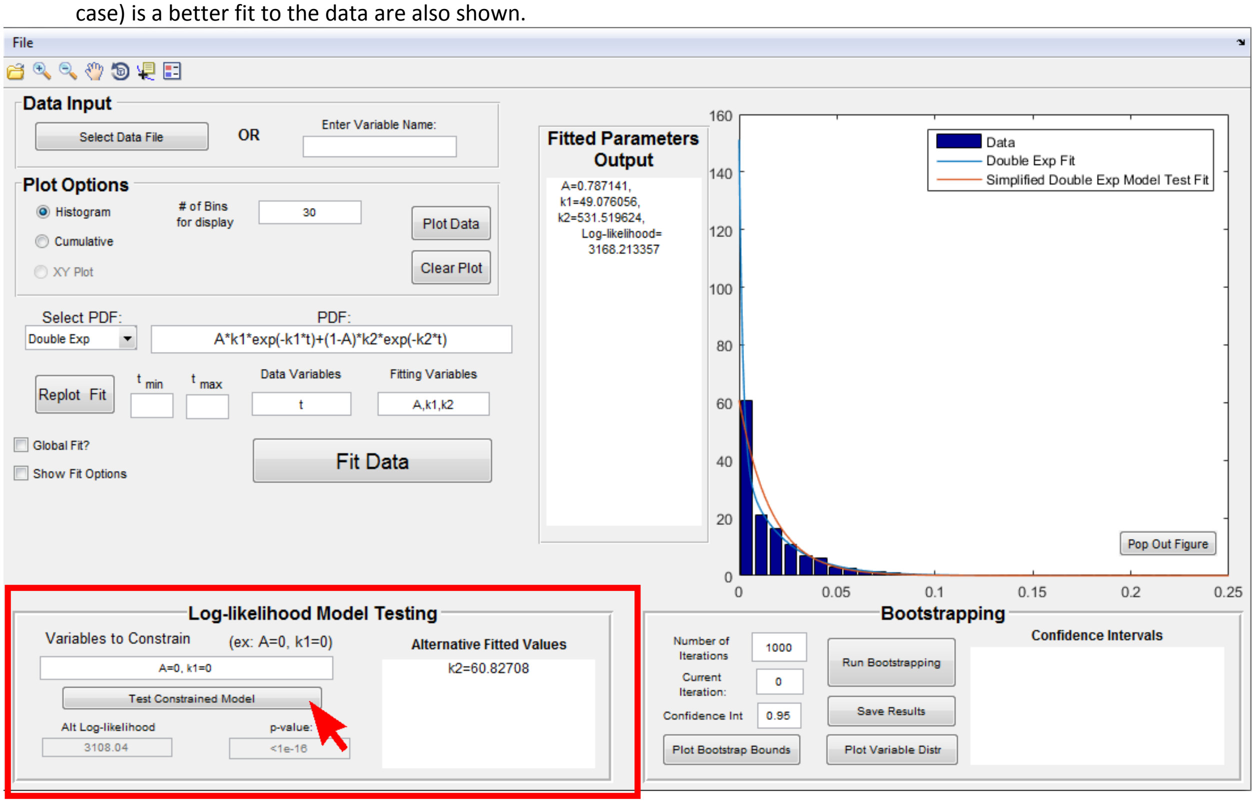


4. Click *Fit Data*
5. Observe the fitted values and compare to the values used in the simulation to produce the data (k_0=20/, d=1.5nm)



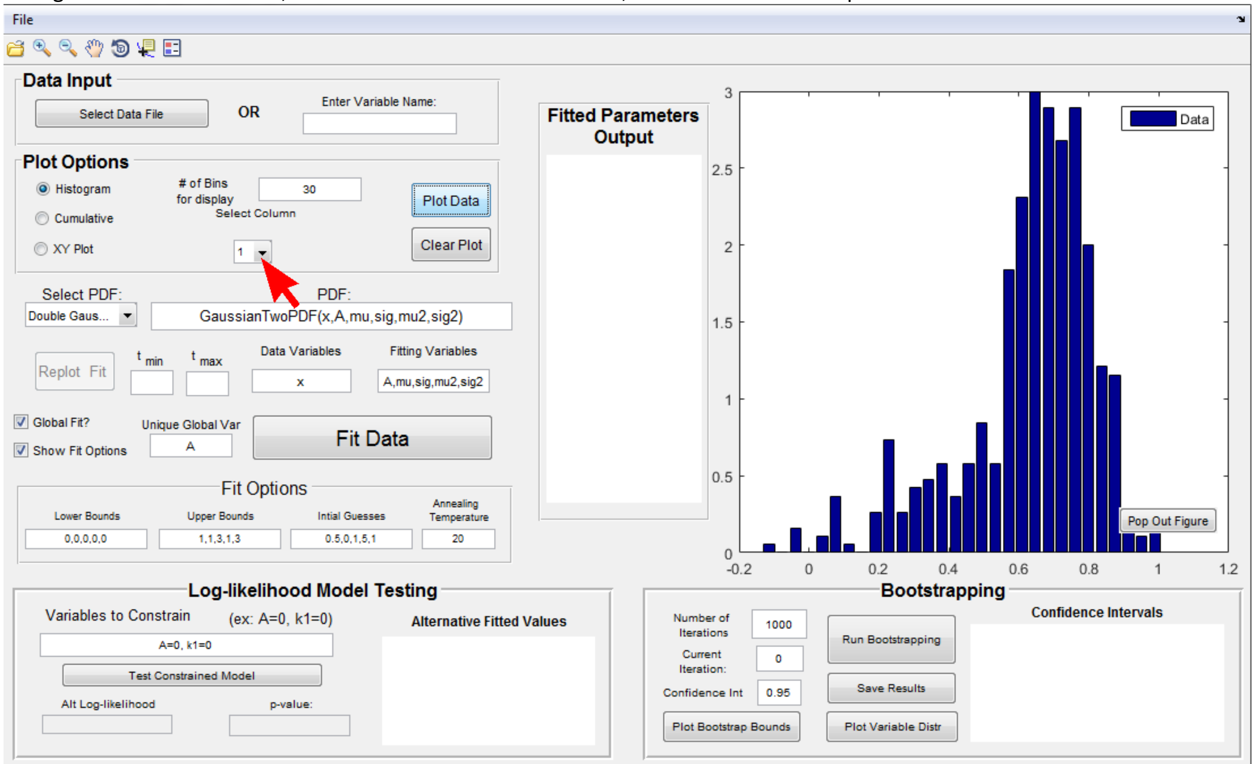
Tutorial 4: Performing Log-likelihood Model Testing

1. Perform all three steps from tutorial 1
2. Specify fixed variables for log likelihood testing
   a. Ensure "Double Exp" is selected from the PDF List
   b. Ensure the *Variables to Constrain* box contains "A=0, k1=0". This constrains the double exponential PDF to effectively become a single exponential PDF with only one rate, k2.
3. Perform the constrained fit by clicking *Test Constrained Model*
4. Observe results
   a. The *Alternative Fitted Values* will give the fitted values of the variables not constrained in the Constrained Model (in this case, the fitted value of k2)
   b. The log-likelihood and the p-value corresponding to likelihood that the unconstrained model (double exponential in this case) is a better fit to the data are also shown.

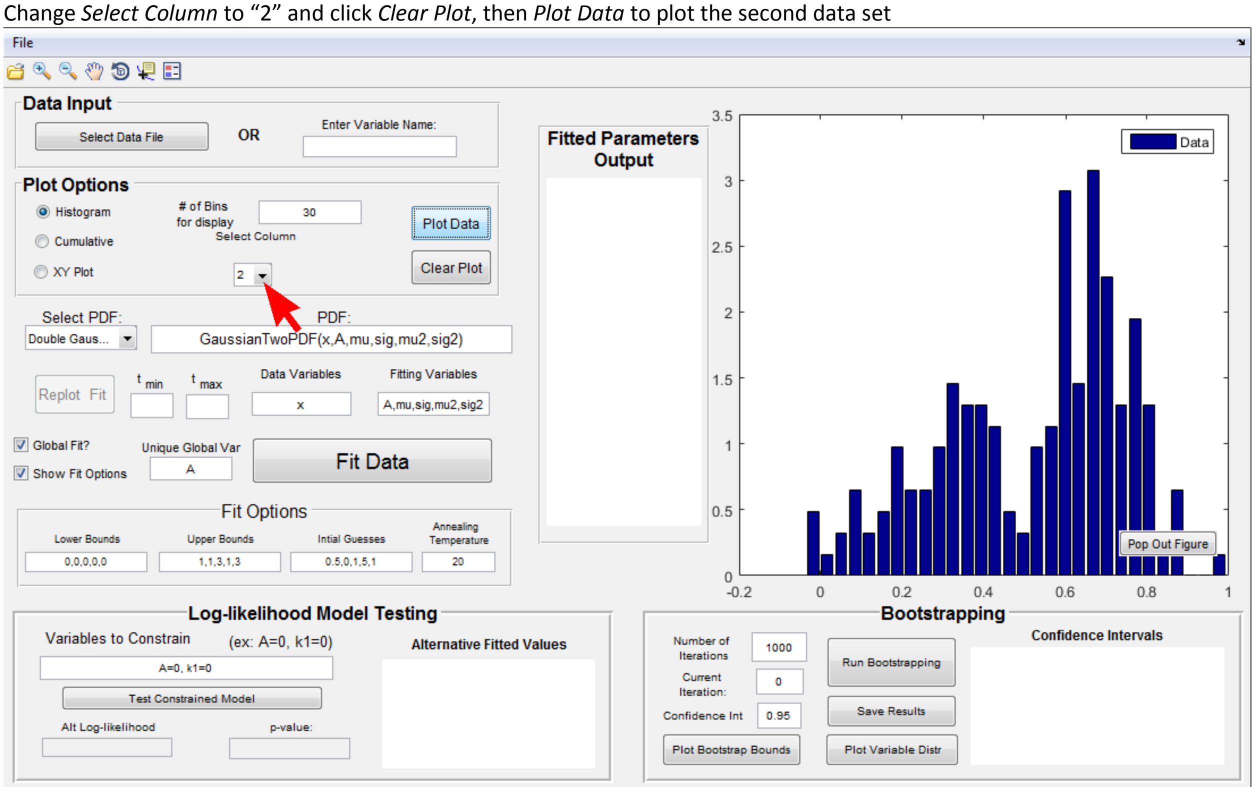


Tutorial 5: Performing Global Fits

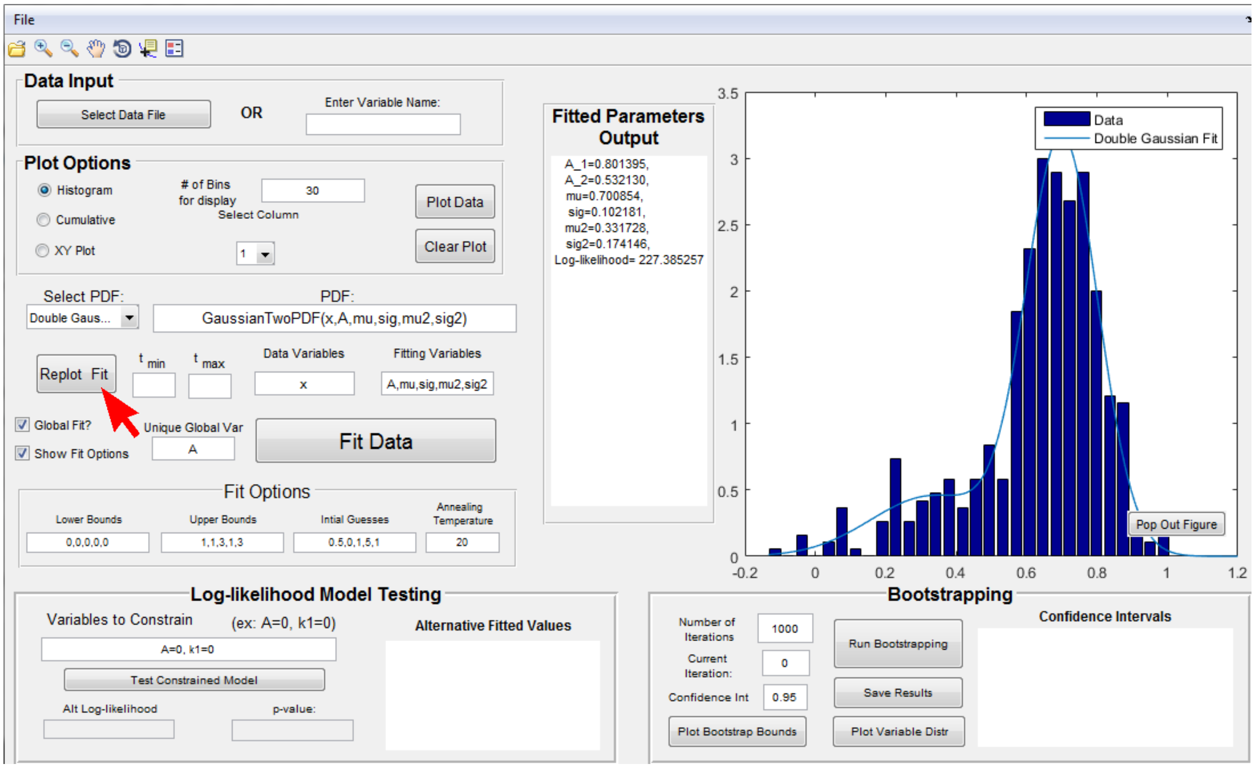
1. Load the Global Example Dataset
2. Select a histogram plot with 30 bins.
3. Change *Select Column* to "1", make sure the tmin box is cleared, and click *Plot Data* to plot the first data set



4. Change *Select Column* to "2" and click *Clear Plot*, then *Plot Data* to plot the second data set



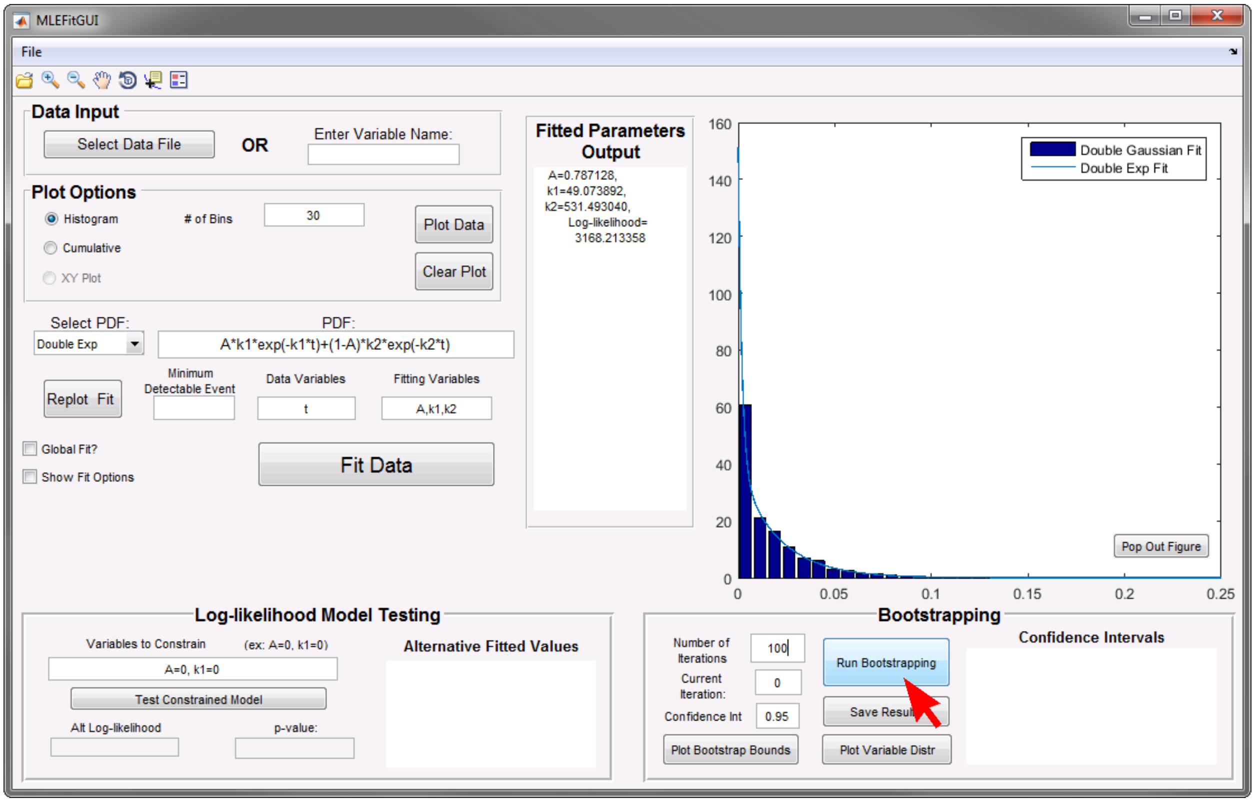
5. Select *Double Gaussian* from the PDF list
6. Check the "*Global Fit?*" Checkbox
7. Enter "A" into the *Unique Global Var* box to allow the amplitude to be fit independently for the two data sets, but for the other fitted parameters to be shared.
8. Click *Show Fit Options*, and change the *Lower Bounds* to 0,0,0,0,0 ; the *Upper Bounds* to 1,1,3,1,3 ; and the *Initial Guesses* to 0.5,0,1,1,1 since this data simulates FRET efficiency which varies from 0 to 1.
9. Click *Fit Data*
10. The parameters are given in the *Fitted Parameters Output* box right with A_1 corresponding to the first data set and A_2 corresponding to the second data set.



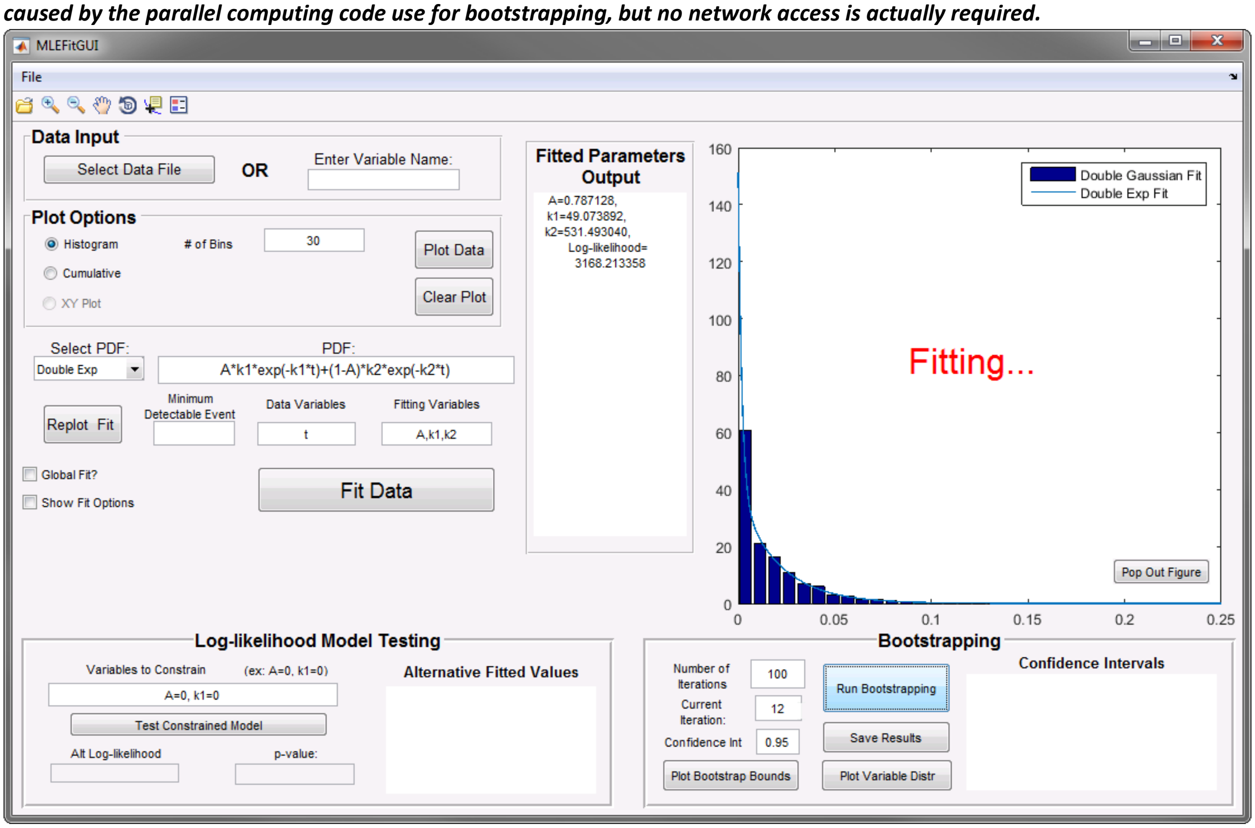
11. To visualize each data set and the corresponding fits, click *Clear Plot*, then use the *Select Column* selector to pick the data set of interest, click *Plot Data*, then *Replot Fit*.

Tutorial 6: Finding Confidence Intervals via Bootstrapping

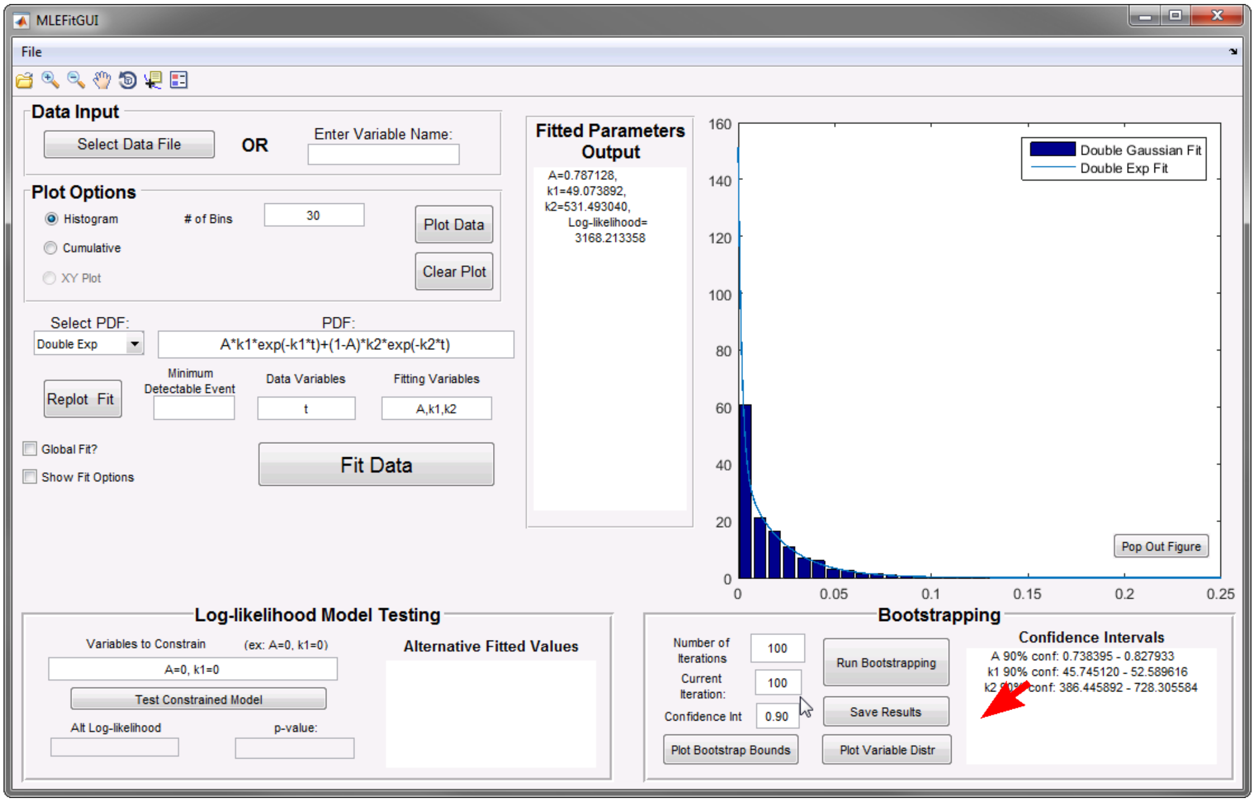
1. Perform all steps listed in Tutorial 1 to get a fit for the double exponential data
2. Input the number of bootstrapping rounds to perform, (for speed, use 100 now, but 500-1000 rounds is recommended)



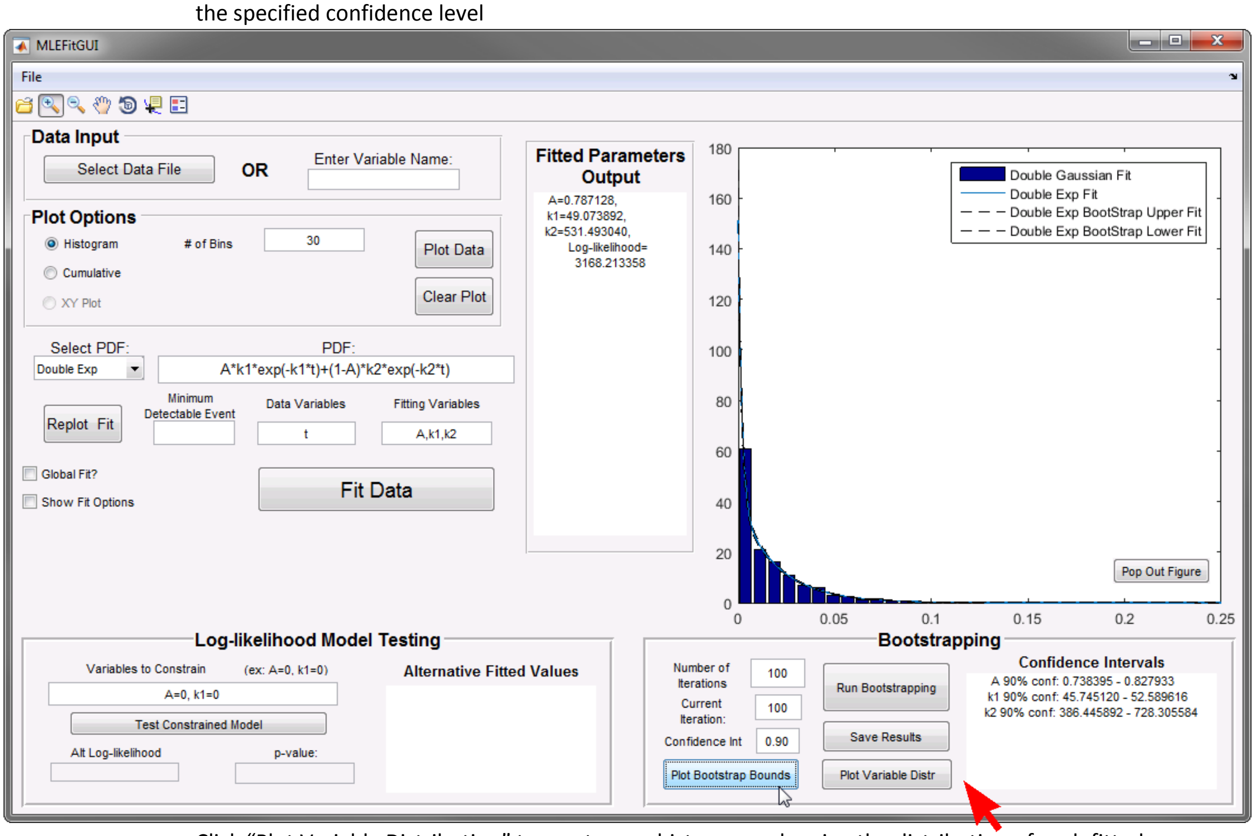
3. Click Run Bootstrapping and wait until the red “Fitting…” text disappears, which will likely take between 1-10 minutes depending on the speed of the computer (the number of current the iteration will increase in increments based on the number of computer cores being used for the computation). ***You may receive a request to allow MEMLET or Matlab to pass through your computer’s firewall. You may deny this request with no loss of functionality. It is caused by the parallel computing code use for bootstrapping, but no network access is actually required.***



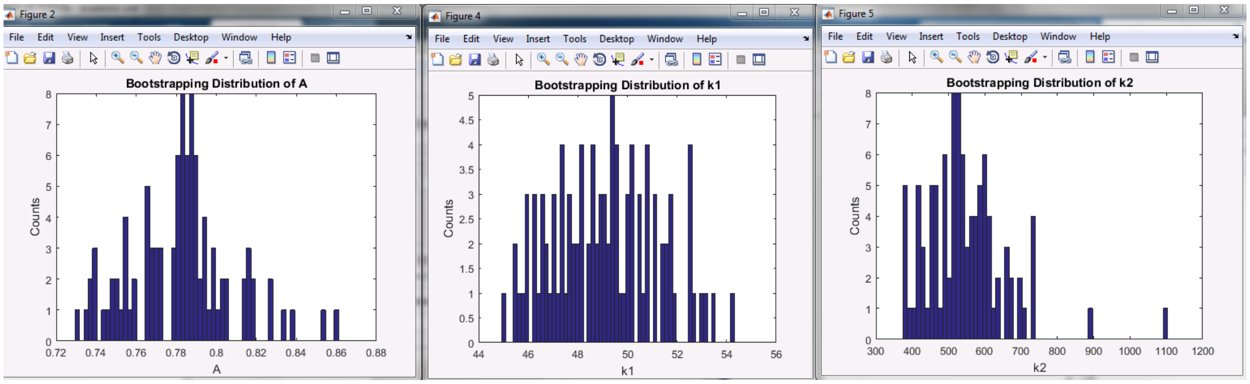
4. Use bootstrapping results
    a. Enter a new confidence level of 0.90 (for 90% confidence intervals, or input or anything between 0 and 1) to see the corresponding confidence intervals in the box in the bottom right.



    b. Click "Plot Bootstrap Bounds" to see the confidence intervals of the PDF plotted on the main display using the specified confidence level



    c. Click "Plot Variable Distribution" to create new histograms showing the distribution of each fitted parameter from the bootstrapping procedure. One window will be created for each fitted variable, but the windows will be created on top of each other, making only one visible until the top window is moved.



d. Click *Save Results* to save all the bootstrapped values in a text file for use outside of the program.