

SCALiR Tutorial

This quick (~5-10 minute) tutorial helps users understand different features of SCALiR and how to visualize and customize data generated by the app. Instructions are also available on the main page of the web-based app.

External standard curves

Step 1. Create a new folder on your device to store the sample data upload files. Open the SCALiR web application and on the left-hand side of the screen, click on the link to the standards concentration file. Repeat this step for the three peaklist data files (two for Mint data and one for Maven data).

NOTE: the “ μ ” symbol denotes “ μ ” in the units column of the standards concentration file.

1) Please upload standards concentrations file

[Click here to download an example of the standards concentrations file](#)

[Click here to download an example of the internal standards concentrations file](#)

Upload standards concentrations file

Drag and drop file here
Limit 200MB per file

Browse files


2) Please upload the peaklist data file. Data from Maven or Mint are accepted. Data files may be in .csv or .xlsx format.

[Click here to download an example of the Mint dense peaklist file](#)

[Click here to download an example of the Mint full peaklist file](#)

[Click here to download an example of the Maven peaklist file](#)

[Click here to download an example of the](#)



An APP for computing concentrations using standard curves. V1.0.0

check out this [link](#) for the source code

Click here to see instructions

Close

or download the tutorial for better explanation

Close

Please upload your standards concentrations file to start

No peaklist datafile has been uploaded

There are no results to show

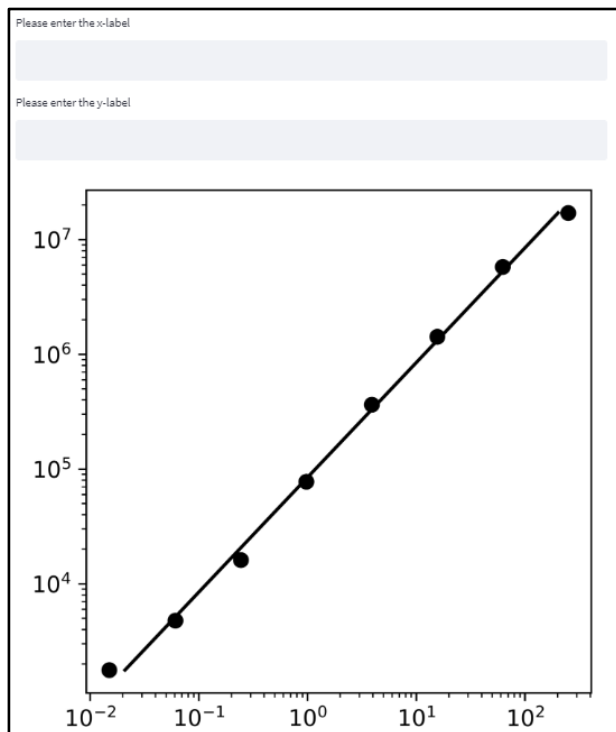
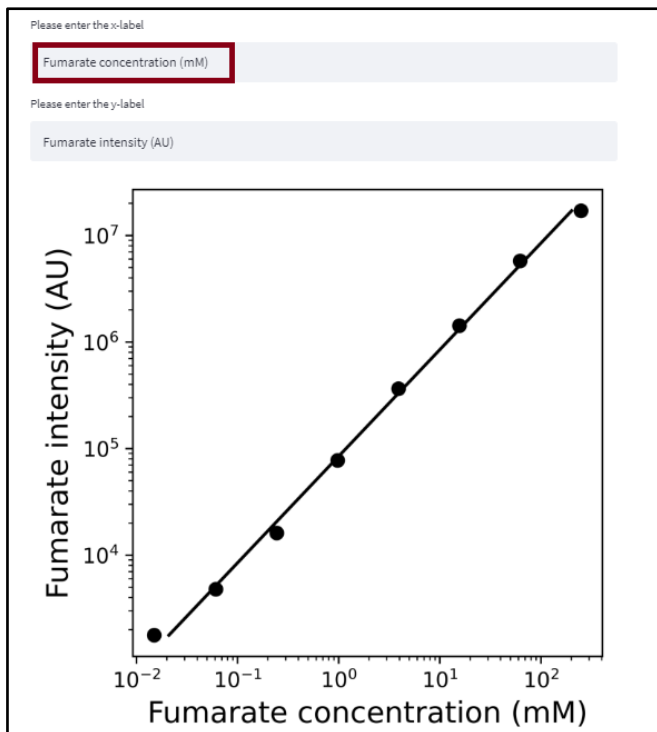
Step 2. On the web application, upload the standards concentration file and the “SCALiR_Maven_Peaklist_sample” file. The data will now pop up on the screen. Scroll down and select “Maven” in the program selection menu. The default option for slope fitting is “Fixed fit” and you should see the links to the standard curve parameters and concentration datafiles pop up below.

The screenshot shows the SCALiR web application interface. On the left, there is a sidebar with instructions: "2) Please upload the peaklist data file. Data from Maven or Mint are accepted. Data files may be in .csv or .xlsx format." Below this are four links to download example files: "Mint dense peaklist file", "Mint full peaklist file", "Maven peaklist file", and "internal standards peaklist file". There is also a file upload section with a "Browse files" button. The main area shows a file named "SCALiR_Standards_Conce..." (0.7KB) and a file named "SCALiR_Maven_Peaklist_s..." (2.4KB). A message states "Data uploading or parameter settings incomplete" and "There are no results to show". A dropdown menu for "Select the program used for generating the peaklist data" is open, showing "Mint" and "Maven" options, with "Maven" selected. Below this is a dropdown for "intensity measurement" with "peak_max" selected.

Step 3. You can click on the highlighted links to download these files. Scroll down to the bottom of the page. The app is displaying the default metabolite. Click on the compound menu and select “Fumarate”. If necessary, you can now manually change the axis labels on the displayed log-log graph from the units you provided in the concentration upload file.

The screenshot shows the "The standard curves have been fitted" section of the SCALiR web application. It includes three links: "Click here to display/hide the standard curve parameters table", "Click here to download your standard curve parameters table" (highlighted with a red box), "Click here to display/hide the concentration data table", and "Click here to download your concentration data" (highlighted with a red box). Below these links is a "Log-log plot visualization:" section. It contains a description: "Description: This plot shows the x- and y-axes in the log scale, with the axis tic". There is a dropdown menu for "select the compound D-Glucose 6-Phosphate will be used by default" with "Fumarate" selected (highlighted with a red box). Below this are two input fields: "Please enter the x-label" with "Fumarate concentration (μM)" and "Please enter the y-label" with "Fumarate intensity (AU)".

Step 4. Change the x-axis label to “Fumarate concentration (mM)” and click enter. If you would like to leave the axis labels blank, delete all text in the box and click enter. To download the plot, right-click and select “Save as.”.



Step 5. Refresh the browser. Upload the sample standards concentrations and the “SCALiR_MINT Peaklist_Full_Results_sample” files. The default program and table type are Mint and full results, so you do not need to change those but can select “peak_area” under the intensity measurement menu. Also, select “Interval fit” under the line of best fit menu. Change the slider bar to 0.90 – 1.10 from the recommended interval of 0.85 to 1.15. SCALiR will re-run the algorithm and your new results will pop up below as before so you can download the data files as well as standard curve visualizations. To use the “SCALiR_MINT_Peaklist_Dense_Peak_Max_sample” file, select “dense peak_max” for the type of table used (peak max is the only intensity measurement available for this file type).

Select the program used for generating the peaklist data

Mint

Indicate the type of table used, see Mint documentation for details

full results

Please select the intensity measurement.. peak_max will be used as the default value
intensity measurement

peak_area

Select the flexibility for your line of best fit

Interval fit – bounds for slope values can be defined. The interval 0.85-1.15 is recommended

interval
Select a range of values

0.90

1.10

0.00

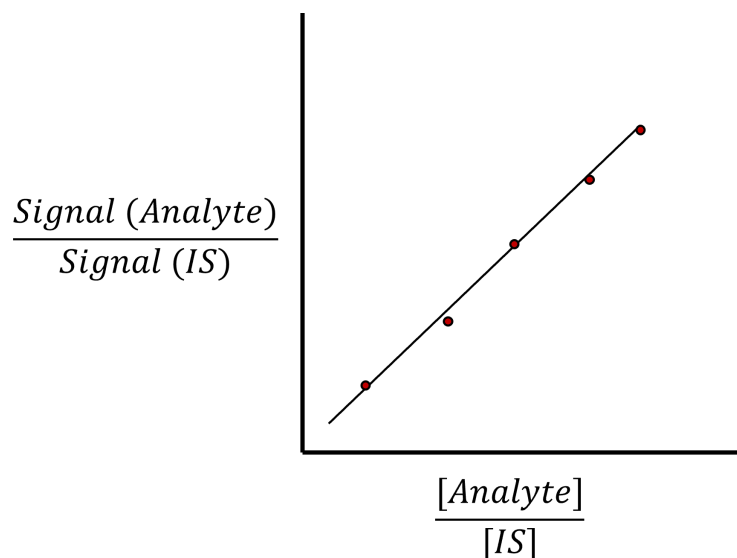
2.00

	0
0	0.9000
1	1.1000

Internal standard curves

In the case where matrix effects are a concern for the analysis, users may wish to spike both their standards and samples with a set concentration of an internal standard (IS). This is usually an isotope labeled version of the analyte of interest.

Users can then upload data generated from taking the ratio of the concentration of the analyte to the concentration of the IS for the internal standards concentration file. For the peaklist data file, users upload the response ratio of the signal intensity of the analyte to the signal intensity of the IS. This will result in a standard curve with the concentration ratio on the x-axis and response ratio on the y-axis:



SCALiR uses the same algorithm as for external standard curves to calculate the line of best fit through the data provided as concentration ratios and response ratios, but uses one additional step to calculate the analyte concentration using the IS concentration. The concentration of the analyte can be solved by multiplying the value of “x” from the line of best fit equation by the concentration of the IS. The user uploads the concentration of the IS in the internal standard concentration file, so the final concentration of the analyte is calculated automatically by SCALiR:

$$\text{Line of best fit equation: } y = m * x + b \quad (1)$$

$$\text{Concentration of analyte: } [\text{Analyte}] = x * [\text{IS}] \quad (2)$$

When users upload the internal standard concentration file and internal standard peaklist file, the data are automatically processed. Users can then choose the fitting flexibility, download standard curve parameters and concentration data files, manually change the log-log plot axis units if necessary, and visualize/download the log-log plots for specified analytes.

The standard curves have been fitted

Click here to display/hide the standard curve parameters table

Click here to download your standard curve parameters table

Click here to display/hide the concentration data table

Click here to download your concentration data

Log-log plot visualization:

Description: This plot shows the x- and y-axes in the log scale, with the axis tic

select the compound Aspartate will be used by default

Aspartate

Please enter the x-label

Aspartate concentration (mM)

Please enter the y-label

Aspartate response ratio

