

Tutorials and worked examples for simulation, curve fitting, statistical analysis, and plotting. http://www.simfit.org.uk

Frequently different scales are required for the x or y axes and it is convenient to make a plot with, say two y axes. For instance, in column chromatography, with absorbance at 280nm representing protein concentration, at the same time as enzyme activity eluted, and the pH gradient. The following table is typical where absorbance could require a scale of zero to unity, while enzyme activity uses a scale of zero to eight, and pH could be on a scale of six to eight.

Fraction Number	Absorbance	Enzyme Activity	Buffer pH
1	0.0	0.1	6.0
2	0.1	0.3	6.0
3	1.0	0.2	6.0
4	0.9	0.6	6.0
5	0.5	0.1	6.2
6	0.3	0.8	6.7
7	0.1	1.5	7.0
8	0.3	6.3	7.0
9	0.4	8.0	7.0
10	0.2	5.5	7.0
11	0.1	2.0	7.2
12	0.1	1.5	7.5
13	0.3	0.5	7.5
14	0.6	1.0	7.5
15	0.9	0.5	7.5

If absorbance and activity were plotted on the same scale the plot would be dominated by activity, so you could change the units of enzyme activity to be compatible with the absorbance scale. However, to illustrate how to create a double graph, a plot with absorbance on the left hand axis and enzyme activity and pH together on the right hand axis will be constructed. Obviously this requires three separate objects, i.e., files for program **simplot**, and information to indicate which y axis is to be used for the individual files. For instance, you could create the following files using program **makmat**, and the data in the above table. File 1: The first column together with the second column (as in test file plot2.tf1)

File 2: The first column together with the third column (as in test file plot2.tf2)

File 3: The first column together with the fourth column (as in test file plot2.tf3)

Then select program **simplot** and choose to make a double graph. Input the first file (absorbance against fraction) scaled to the left hand axis with the other two scaled to the right hand axis to get the first figure following. To transform the default **simplot** plot into the finished product of the second figure following proceed as follows:

- a) Edit the overall plot title and both plot legends.
- b) Edit the data ranges, notation and offset on the axes.
- c) Edit the three symbol and line types corresponding to the three files.
- d) Include an information panel and edit the corresponding keys.
- e) Create the final PostScript file.

Original x,y Coordinates _[8.00 ר 1.00 0 Δ 0 \Box Δ 0.75 0 0 0 0 6.03 Δ E-xtra axis 0 0.50 -4.05 0 > 0 0 0 0 0.25 -2.08 Δ 0 Δ Δ Δ 0 0 0 0 Δ Δ Δ $\begin{array}{c} & \\ - \\ - \\ 15.0 \end{array}$ 0.00 4.5 1.0 8.0 11.5 Х



