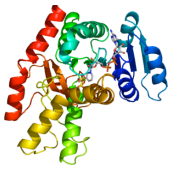
# Biochemistry Lab
## *Preparing Figures and Figure Legends*



**Assignment** – 1) 25 pts: Prepare one gel or Western blot from ANY of your work conducted this semester WITH a figure legend. 2) 25 pts: Prepare one graph from your work. Include a proper figure legend.

**General Guidelines:**
A nice set of figure guidelines is included on the "Resources for Authors" page of the Journal of Biological Chemistry. http://www.jbc.org/site/misc/itoa.xhtml
Although focused on images for digital submission, many of the tips are generally applicable to any figure preparation. http://art.cadmus.com/da/jbc/guidelines.html In particular the sections on font usage and multi-panel figures are particularly helpful.

*Note*: JBC recommends against assembling figures in PowerPoint because of potential loss of color depth and resolution, **however**, this can be a very good way to assemble your figure for our purposes as it can be arranged as desired and then saved as a single file. In particular this will be a nice way to prepare individual slides that you can then print out and pin up to create your poster presentation. Once prepared, you could save the image as a png or jpg file for importing into any document.

**Figure Legends**: Please note that figure legends can include both simple conclusions of the data and are a mini-method section. Carefully read these examples and other examples from the journal articles you have collected. You are expected to write at the same professional and formal level (including the style and detail) as these legends.

**Figures:**

- Pictures of gels, blots, or other figures must be completely labeled so the reader can understand the data you are presenting. What is in each lane should be indicated in the legend. The molecular weights should also be indicated on the side and any points of interest highlighted by an arrow. See Figure 1A below (parts B and C of the figure and legend were removed for clarity). All examples shown are from this paper: Wang, Q., Yu, L., and Yu, C.A. (2010) *J. Biol. Chem.* **285,** 10408-10414

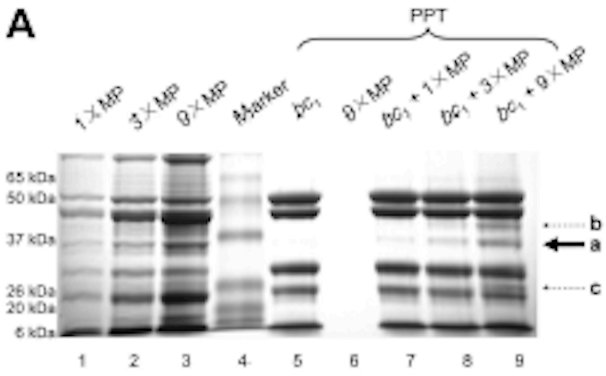
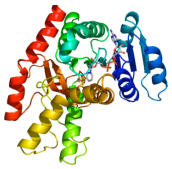



**Figure 1. Identification of MDH as an interacting protein with the $bc_1$ complex.** A, SDS-PAGE analysis of $bc_1$ pulled-down proteins from matrix extract. *Lanes 1-3*, various amounts of MP as controls show the proteins in the matrix extract; *lane 4*, standard proteins; *lanes 5 and 6*, $bc_1$ complex and MP only as controls show the precipitates after centrifugation; *lanes 7-9*, shown is the $bc_1$ complex together with the pulled-down proteins from matrix extract. The *solid arrow* shows the main matrix protein being pulled down by the $bc_1$ complex. The *dashed arrow* shows some other bands being pulled down by the $bc_1$ complex. *MP* stands for mitochondrial matrix proteins; 1X, 3X, and 9X stand for 0.28, 0.83, and 2.50 mg/ml of MP in the experiment. *PPT* stands for precipitates. *a*, malate dehydrogenase; *b*, aspartate aminotransferase; *c*, unknown protein.

Notice how the figure title summarizes the main point of the experiment, but the legend itself provides only the relevant experimental information and observations needed to understand the figure. In this paper, the experimental details are in the "Experimental Procedures" section, while the interpretation of the data is found in the "Results and Discussion" section.

Any new abbreviations were defined as needed. MDH and $bc_1$ had been previously defined at the beginning of the paper.



**Graphs:**

Graphs are to be prepared on the computer. (It doesn't matter which program you use.)
Any graph should follow the following guidelines:

- Each axis must be properly labeled. Do not include a legend within the graph.
- Make the graph fit the data. Limit the axis to the values in the graph.
- When you have multiple data points (n≥1) for a single reading, do NOT graph all points on one graph. Instead, average them. If the number of replicates is three or greater, then determine the standard deviation and include that information in your graph.
- Remove guidelines from the background of the graph, it should look uncluttered so the data points (and trendline, if necessary) are clearly visible.
- A figure number and legend will be found underneath each graph. The title for each graph is in the figure legend, *not at the top of the graph*. Each legend should be a mini statement on what you did to get the graph. It is kind of a specific methods section for each graph.
- Do not include the table of data used to generate the graph unless requested (it is in your notebook!).
- The standard default Excel form is NOT acceptable. Lines through the graph will not be accepted. Adjust the size and font of the axis label and numbers accordingly. The default size and font are typically not appropriate.
- Line graphs should be connected ONLY IF there is some sort of numerical / formulaic relationship between points (time, concentration, etc…). Histographs (bar graphs) are for values that are not related (cell lines, types of treatments, etc…).

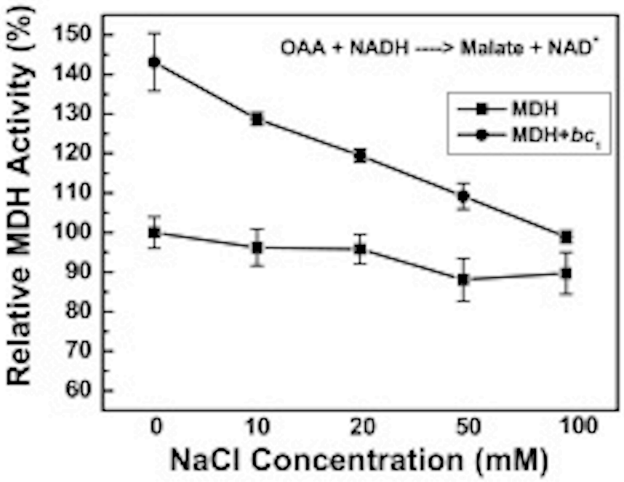



**Figure 6. Effect of ionic strength on reverse MDH activity enhancement by $bc_1$.** MDH only or together with $bc_1$ complex in 100 mM $Na^+/K^+$ phosphate buffer, pH 7.5, was incubated with the indicated concentrations of NaCl on ice for 30 min before MDH activity (the reverse reaction) was determined. The activity of MDH only was used as 100%. n = 4, data are the means ± S.D.

**Tables:**

If there are many graphs necessary for a set of data, such compiled data can be presented in a detailed table. Often one representative graph will be included in a figure and sometimes the remaining associated figure(s) will be published as supplementary data.

**TABLE 3**

**Kinetics analysis of the MDH in the present of $bc_1$ complex**

0.1, 0.05, 0.025, 0.016, and 0.0125 mM NADH and 0.2 mM OAA were used in the assay for analyzing the $K_m$ of NADH. 0.1, 0.025, 0.0125, and 0.00625 mM OAA and 0.2 mM NADH were used in the assay for analyzing the $K_m$ of OAA. MDH-only or together with $bc_1$ complex was used in both of the assays, and $V_{max}$ was also calculated via this Lineweaver-Burk plot.

| Enzyme | NADH | | OAA | |
|---|---|---|---|---|
| | $K_m$ | $V_{max}$ | $K_m$ | $V_{max}$ |
| | μM | *mmol/min/mg* | μM | *mmol/min/mg* |
| MDH | 22.2 ± 0.5 | 1.6 ± 0.2 | 10.7 ± 0.7 | 2.2 ± 0.1 |
| MDH + $bc_1$ | 15.4 ± 0.5 | 2.5 ± 0.2 | 8.9 ± 0.9 | 3.5 ± 0.1 |