

Week 1 Bio375 Lab Agenda

1. Assign groups
2. Set up work area
2. Review lab policies and syllabus
4. Pipet Olympics (practice dilution calculations and pipetting)
5. Discuss Blood Cells lab (intro)
6. Sign up for paper presentations
7. Discuss writing rules



Assign Lab Groups

Group number	Student	Group number	Student
1		4	
2		5	
3		6	

Set up work area

- Get oriented with the lab. Locate gloves, pipet tips, microfuge tubes, tube racks, glass disposal container.
- Label tape with group # and your names and stick it on top-most drawer. This drawer should contain:
 - Beaker with 1.5 mL microfuge tubes
 - 3 pipetmen - 1 mL, 200 μ L, 20 μ L
 - One box each of large and small pipet tips
 - Permanent markers
 - Microfuge tube racks (large yellow; small white)
 - Long green test tube rack
 - Tape
- Center isle of each station should have large beaker for discarding experimental trash.

Lab policies

- Lab **attendance is mandatory** and there will be no make-up lab. If you need to miss lab due to illness, religious holidays, other special/unforeseen circumstances, please contact me as soon as possible. In case you miss a lab day, it is your responsibility to obtain data from group mates to complete your lab report.
- It is very important to **arrive to lab on time (1:10 pm)**. Lab classes will start with me providing important instructions and tips which will help you conduct your experiments efficiently.
- No eating or drinking in the lab; wear closed-toed shoes; long hair **MUST** be tied up.

Lab manuals, notebooks, reports

- **Lab manuals** posted on Moodle. Read *prior* to lab and complete pre-lab calculations/preparations before-hand. Ask questions if unsure of anything.
- **Lab notebook.** Each group member must acquire and record all necessary information and data in their own individual notebook. In the research notebook, you should record:
 - Design and specific goals
 - Thoughts & questions
 - Observations and data
 - Changes made to the protocol
 - Conclusions/interpretations from your results
- **Four lab reports. (hard copies).** Details on how to write these lab reports will be provided. Although you will be working in groups, all written work submitted must be your own.

BIO 375: Biochemistry **Fall 2026 Lab Syllabus** **(subject to change)** Lab manuals posted on Moodle



Date	LAB EXERCISE
Sept. 2	NO LAB
Sept. 9	• Getting to know each other and our lab space (read lab policies) • Pipet Olympics (read appendices 1 and 2) • Learning how to write scientific papers • Introduction to Blood Cells Lab (please read introduction in posted manual)
Sept. 16	• Blood Cells Lab, Part I: Extract proteins
Sept. 23	• Viewing protein structures in 3D – myoglobin (please download <u>PyMol</u> on your laptop and bring to lab)
Sept. 30	• Blood Cells Lab, Part II: Protein assays
Oct. 7	• Blood Cells Lab, Part III: SDS-PAGE analysis • Introduction to Enzyme Kinetics Lab (please read introduction in posted manual) <i>(Blood Cells lab report due Wed, 10/21)</i>
Oct. 14	FALL BREAK – NO LAB
Oct. 21	Enzyme Kinetics Lab, Part I: Determining the effect of substrate concentration

Pipetting

- Please choose the **correct pipetman** and set it to:
 - 976 μL
 - 154 μL
 - 15.5 μL
 - 2 μL

Pipet Olympics

- Reinforce proper pipetting technique.
- Practice calculating and making dilutions.
- Use data collected to create a scatterplot and fit a least squares regression line *in Microsoft Excel – please do NOT use Google excel.*

Blood cells lab intro

(refer to lab manual)

Calculations (pg. 7 Blood cells lab manual)

1. For the BSA standard curve, set up **six eppendorf tubes** and label appropriately. Add BSA standard stock solution (1 mg/mL) to obtain the final concentrations shown below. Final volume will be 100 μ L. (Use $C_1V_1=C_2V_2$ to calculate V_1 of BSA stock).

Final concentration (mg/mL)	μ L of 1 mg/mL BSA to add	μ L of water to get to 100 μ L
0	0	100
0.2		
0.4		
0.6		
0.8		
1	100	0

Calculations (pg. 7 Blood cells lab manual)

Since you have no idea how concentrated/dilute your samples are, you will need to assay different dilutions of your sample to cover a range of concentrations, to ensure that at least one dilution “hits” the linear range of the BSA protein standard curve. We will try three dilutions per sample: undiluted, 1:10 diluted, 1:100 diluted. Prepare your 1:10 and 1:100 dilutions in **eppendorf tubes in 100 μ L final volume of water. Label tubes appropriately, so you don’t mix them up!**

Dilution	μ L of sample to add	μ L of water to get to 100 μ L
1:10		
1:100		

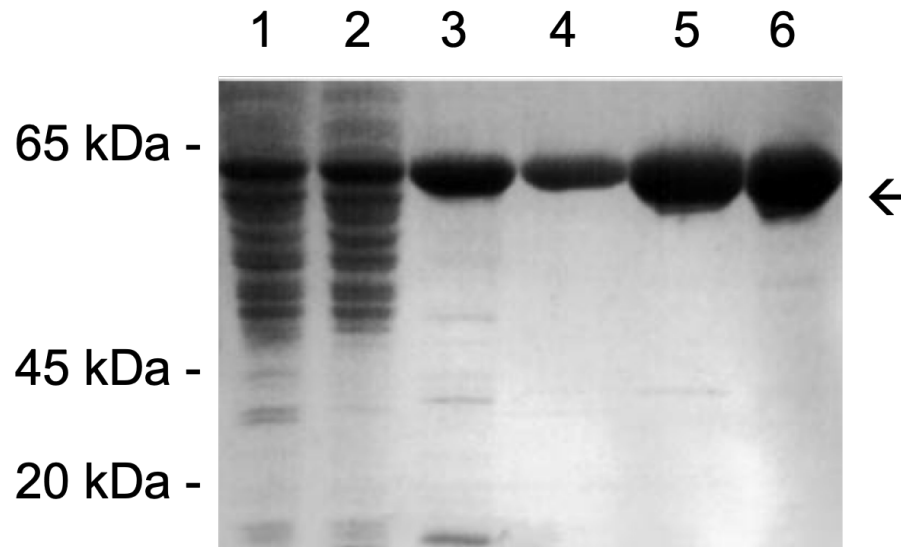
Sign up for paper presentations

Writing rules - Figures

(pp 15-16 blood cell lab manual)

Figures

The figure should be numbered, include a title, and include a detailed explanatory caption (figure legend) **below** the figure. Here is an example:



Writing rules - Figures

(pp 15-16 blood cell lab manual)

Figures

The figure should be numbered, include a title, and include a detailed explanatory caption (figure legend) **below** the figure. Here is an example:

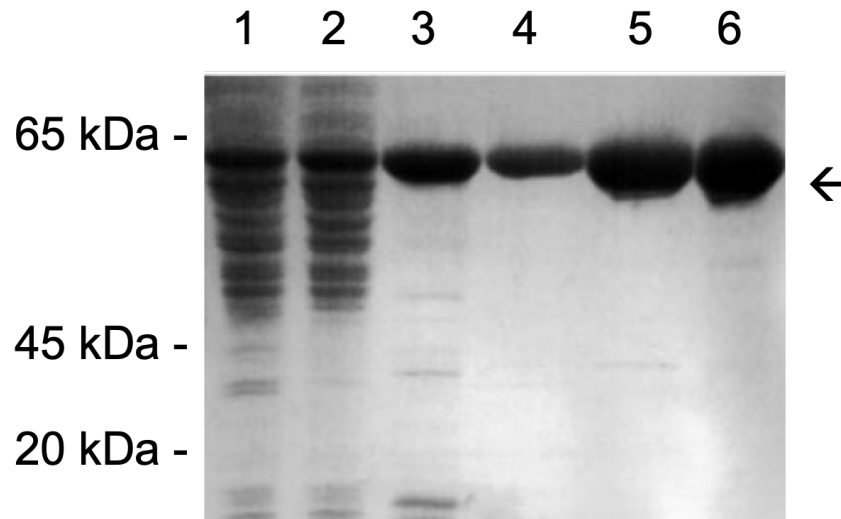


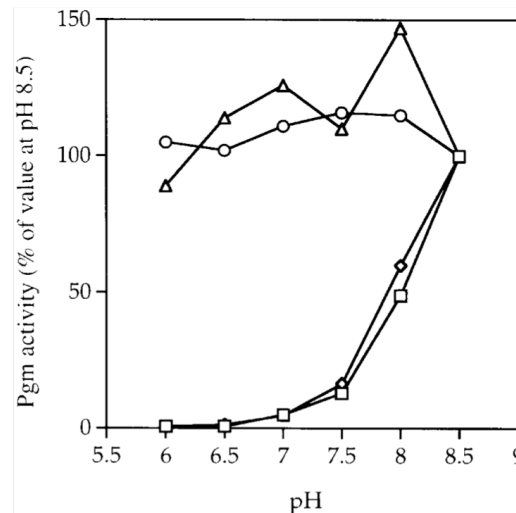
FIG. 2. SDS-PAGE of *B. stearothermophilus* PGM. SDS-PAGE of samples taken throughout the purification of methionyl PGM was carried out under reducing conditions as described under Materials and Methods, and the gel was stained with Coomassie blue. 5 μ g of protein was run in the various lanes except for lane 4, which had only 3.5 μ g. The samples in the various lanes are from (1) crude lysate supernatant fluid; (2) dialyzed ammonium sulfate precipitate; (3) heat step supernatant fluid; (4) DEAE-Sephadex column pool; (5) octyl-agarose column pool; and (6) S200 column pool. The numbers to the left of the figure indicate the position of molecular mass markers in kDa run on the same gel, and the arrow denotes the PGM.

Writing rules - Graphs

(pp 15-16 blood cell lab manual)

Graphs

- A graph is also a figure and should be numbered and labeled according to the rules for Figures.
- Axes should be labeled and include units of measurement
- Tick marks on both axes should be at intervals frequent enough to allow reader to estimate value of each data point
- The meaning of each symbol should be clearly indicated
- Here is an example:



Writing rules - Graphs

(pp 15-16 blood cell lab manual)

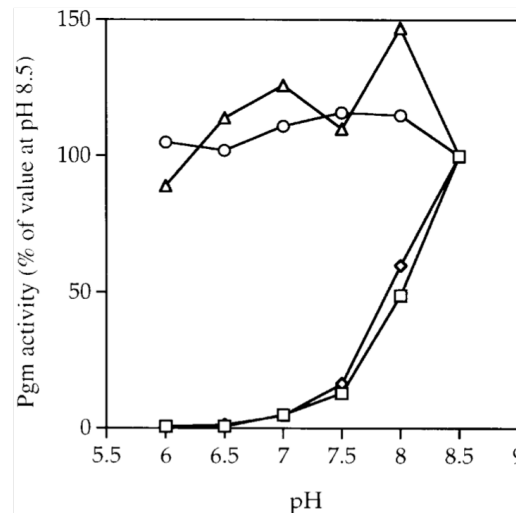


Fig. 1. The response of Pgms to pH. The Pgms from *C. perfringens* (□) and *Staphylococcus saprophyticus* (○) were treated with EDTA prior to assaying by the two-step assay with 10 μM total Mn^{2+} at various pHs. The Pgms from *M. luteus* (□) and *Streptomyces coelicolor* (Δ) were assayed at the same pHs in the absence of metals with 1 mM DPG. Pgm activity is reported as a percentage of the activity obtained at pH 8.5.

Writing rules - Tables

(pp 15-16 blood cell lab manual)

Tables

Tables should be numbered and have a short, but informative title **above** the table. Any details that you want to highlight can be included as a footnote to the table. Here is an example:

pH 6.0		pH 7.0		pH 8.0	
Co ²⁺ concn.	Pgm activity	Co ²⁺ concn.	Pgm activity	Co ²⁺ concn.	Pgm activity
None	<0.1	None	<0.2	None	<0.1
7.5 μ M	2	3.2 μ M	11	2.7 μ M	28
77 μ M	7.6	32 μ M	33	28 μ M	61
1 mM	11	0.4 mM	52	0.3 mM	100

Writing rules - Tables

(pp 15-16 blood cell lab manual)

Tables

Tables should be numbered and have a short, but informative title **above** the table. Any detail that you want to highlight can be included as a footnote to the table. Here is an example:

Table 3. The pH dependence of *C. perfringens* Pgm in the presence of Co^{2+} .

pH 6.0		pH 7.0		pH 8.0	
Co^{2+} concn.	Pgm activity	Co^{2+} concn.	Pgm activity	Co^{2+} concn.	Pgm activity
None	<0.1	None	<0.2	None	<0.1
7.5 μM	2	3.2 μM	11	2.7 μM	28
77 μM	7.6	32 μM	33	28 μM	61
1 mM	11	0.4 mM	52	0.3 mM	100

Notes: Assays were carried out using EDTA-treated Pgm as described in the text. Co^{2+} concentrations given are the free concentrations. PGM activity is reported as a percentage of the activity obtained at pH 8.0 with 0.3 mM free Co^{2+} . This value was set at 100%.