1. (30 pts) Amino acid structure, the peptide bond, and acid-base
   (a; 3 pts) Why is histidine frequently found in protein active sites?

   It is the only AA with a pKa near neutral pH, so it
   frequently is involved in proton transfer.

   It can chelate metal ions like Fe and Zn

   (g; 4 pts) Calculate the ratio between the protonated and deprotonated forms of the histidine side chain
   at pH 7.2. The protonated form has a pKa of 6.04.

   \[
   \begin{align*}
   \text{pH} & = pK_a + \log \left( \frac{[A^-]}{[HA]} \right) \\
   \log \left( \frac{[A^-]}{[HA]} \right) & = \text{pH} - pK_a \\
   \frac{[A^-]}{[HA]} & = 10^{1.16} = 14.5 > 1 \text{ since pH > pK} \\
   \end{align*}
   \]
(b; 14 pts) Draw the tripeptide His-Pro-Val in its predominant ionic form at pH 5, with all of the peptide bonds in the trans conformation. Start from the ring given below. It's there twice in case you need to redraw.

(e; 9 pts) Indicate on your structure the four atoms that define the Φ angle for the proline residue. Assuming that the proline side chain ring is constrained to be flat, estimate the permitted value of Φ. Would your answer be substantially different if the His-Pro peptide bond were cis? Why or why not?
2. (40 pts) Protein Folding
   
   (a; 9 pts) The thermodynamics of protein folding: What are the two main contributors to $\Delta S$, and what are their signs and the sign of the overall $\Delta S$? What is the sign of $\Delta H$? What is the sign of $\Delta G$ for protein folding?

   \( \Delta S \) of conformational restriction is $\Theta$ for folding
   \( \Delta S \) of hydrophobic effect is $\theta$ for $\text{H}_2\text{O}$ release upon collapse
   net $\Delta S$ for folding is $\Theta$ (unfavorable)
   $\Delta H$ is $\Theta$ for noncovalent bond formation
   $\Delta G = \Delta H - T\Delta S$ is typically $\Theta$ at low $T$, $\Theta$ at high $T$ [in some cases the temperature dependence of the hydrophobic effect itself leads to $\Theta \Delta G$ at old $T = \text{"old denaturation"}]$

   (b; 6 pts) Explain why H-bonds and electrostatic interactions make contributions to stability that are quantitatively much smaller than the binding energies of the H-bonds and electrostatic contacts seen in proteins. Why are they still important for the specificity of folding?

   - An H-bond in a protein that would be replaced by H-bonds to water if the protein unfolded. So the increased stability of the protein reflects the difference in the strength of the protein H-bond vs. the water H-bonds. Similarly, the participants in an electrostatic interaction would otherwise be solvated in water.

   - However, in a mutant or a misfold, buried protein moieties lose the interactions with water but do not gain new molecular interactions - this is a net loss and dis-favors all but native-like structures.

   Score for the page 15
Here is a proposed mechanism for the GroEL/ES folding machine.

**Fig. 2.** The polypeptide folding cycle at GroEL. (a) The initial polypeptide acceptor state *in vivo* and in a

(c; 6 pts) We listed two related but distinct functions for the chaperonin. What are they; in other words, what does it mean to chaperone the process of protein folding?

1. Allow repeated cycles of folding → unfold to prevent getting trapped in misfolded states on the rugged energy landscape
2. Enforce unimolecular folding to prevent aggregation

(d; 3 pts) What causes a candidate client protein to stick to GroEL?

A misfolded protein will frequently show patches of hydrophobic surface which bind unspecifically to a hydrophobic patch inside the GroEL cavity.

(e; 5 pts) In the c→d step, the protein is released from binding and is allowed to refold on its own. We called the cavity a particular kind of cage, Name it and describe its function.

The **Hofmayer cage** - provides a hydrophilic surface that repels what should be the inside of the substrate protein and enforces unimolecular folding.

(f; 3 pts) The client protein may need to be unfolded and allowed to refold many times. Why does a cyclic process like this require ATP hydrolysis? [If it didn’t use an external energy source, what would happen?]

One of the five states above must be thermodynamically the most stable, and the difference in energy of an
large wrt. $kT = \text{thermal energy}$. A cycle process would get stuck in an **equilibrium**.

[Third going around the directional way always requires energy input]
(e; 8 pts) Sketch the model that protein aggregation can occur through a combination of steric zipper (=stacked β sheet) formation and domain swapping.

3. (30 pts) Biomolecules and Miscellaneous:

(a; 3 pts) Why do membrane phospholipids have two extended alkyl tail groups? Why not one or three?

1. One → micelles lose the head groups are wider than one tail
2. Three → presumably would be less stable as a bilayer due to exposed hydrophobic surface

(c; 6 pts) Draw a phosphatidylethanolamine (ethanolamine = –OCH₂CH₃NH₃⁺) with one saturated R group and one monounsaturated R group with a cis double bond.
(e; 6 pts) Here is the Fischer projection of D-sorbose. Indicate which hydroxyl attacks the ketone to make the furanose form of the ring, and draw the Haworth projection of the furanose ring. Indicate the anomeric carbon stereochemistry with a squiggle.

D-Sorbose

Here is the structure of cellobiose, a disaccharide derived from cellulose.

(3 pts) Circle and name the linkage between the two glucose moieties.

\[
\beta(1\rightarrow4) \text{ glycosidic bond}
\]

Score for the page 9
(12 pts) We discussed several ligands for Hemoglobin, including CO₂, H⁺, and Cl⁻. Explain why it makes sense in terms of physiology for each of them to decrease the binding affinity of Hb for O₂.

**CO₂** - An increase in CO₂ indicates more active metabolism — means more O₂ is needed. Also an indirect indicator of pH and a waste product that must be transported out of the body.

**H⁺** - A decrease in pH is another indicator of more metabolic acid that must also be transported, partially on Hb.

**Cl⁻** - Flows into RBC as H₂CO₃ flows out — again, Cl⁻ is an indirect sensor for metabolic metabolism.