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

   (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.
(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 $\Phi$ angle for the proline residue. Assuming that the proline side chain ring is constrained to be flat, estimate the permitted value of $\Phi$. 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?

(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?
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 cycling reaction in vitro is an asymmetric, ADP-containing complex, as shown in Fig. 1 (Rye et al. 1997; Xu et al. 1997). (b) ATP and non-native polypeptide (green) bind rapidly to the open ring of the ADP asymmetric complex, with ATP producing downward rigid body movement of the intermediate domain via contacts with the nucleotide pocket (Chaudhry et al. 2003) and attendant small elevation and counterclockwise twist of the apical domains (Ranson et al. 2001), the latter of which directly enable GroES binding. ATP binding also sends an allosteric signal to the opposite (trans) ring that ejects its ligands: GroES, substrate polypeptide, and ADP (Rye et al. 1997). The binding of non-native polypeptide along with ATP accelerates such departure of GroES (Rye et al. 1999). (c) Initial GroES interaction with the ATP–polypeptide-bound ring, occurring by y0/C12 s (Ciff et al. 2006), is followed over y0/C18 s by large and forceful rigid body elevation and clockwise twisting movements (60° and 120°, respectively) that eject polypeptide into a GroES-encapsulated hydrophilic chamber where polypeptide attempts to fold (Weissman et al. 1996; Xu et al. 1997; Rye et al. 1997). This folding-active cis complex is the longest-lived state in the folding cycle. (d) ATP hydrolysis, occurring with a half-time of y10 s, primes the cis complex for discharge of its ligands by weakening afinity of GroEL for GroES (Rye et al. 1997). (e) ATP hydrolysis gates the binding of ATP and non-native polypeptide to the trans ring (Rye et al. 1999), a function of allosteric behavior. In the case of ATP, such behavior is anti-cooperative between rings (sequential, KNF) and positive (concerted, MWC) within the rings, overall a 'nested' behavior (Yifrach & Horovitz, 1995). ATP (and polypeptide) binding to the trans ring discharges GroES and polypeptide from the old cis ring and leads to a new round of cis encapsulation and folding in this (opposite) ring. Note that the 10 s or so of folding time in the cis complex is not sufficient for most molecules of stringent, GroEL–GroES-dependent substrate proteins to fold to native form; typically, only a few percent reach the native state. The other molecules are discharged at (e) in non-native states that, in vitro, are rebound by other GroEL complexes for another attempt at folding. In the cell, such non-native forms can also partition to other chaperones or the degradative apparatus, their fate depending on the relative concentrations and afinity of the various components. D indicates ADP, T indicates ATP.

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

(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.

(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?]
(e; 8 pts) Sketch the model that protein aggregation can occur through a combination of steric zipper (=stacked $\beta$ 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?

   (c; 6 pts) Draw a phosphatidylethanolamine (ethanolamine = $-\text{OCH}_2\text{CH}_2\text{NH}_3^+$) 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.

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

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