Biochemistry 463, Summer II **Your Name:** Key

University of Maryland, College Park **Your SID #:**

Biochemistry and Physiology **Prof. Jason Kahn**

Exam I (100 points total) **July 28, 2008**

You have 80 minutes for this exam. (57) → 58

Exams written in pencil or erasable ink will not be re-graded under any circumstances.

Explanations should be concise and clear. I have given you more space than you should need. There is extra space on the last page if you need it.

You will need a calculator for this exam. No other study aids or materials are permitted.

Generous partial credit will be given, *i.e.*, if you don't know, guess.

Useful Equations:

$\Delta S_{system} - \Delta H_{system}/T \geq 0$ $\quad pH = -\log([H^+])$ $\quad E = mc^2$

$S = k\ln W$ $\quad \Delta G = \Delta H - T\Delta S$ $\quad pH = pK_a + \log([A^-]/[HA])$

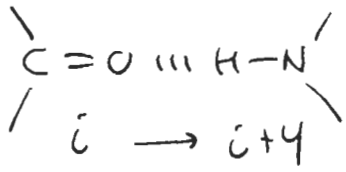
$K_a = [H^+][A^-]/[HA]$ $\quad \Delta G^\circ = -RT\ln K_{eq}$ $\quad e^{i\pi} + 1 = 0$

Honor Pledge: At the end of the examination time , please write out the following sentence and sign it, or talk to me about it:

"I pledge on my honor that I have not given or received any unauthorized assistance on this examination."

1. (23 pts) Amino acid structure and the peptide bond

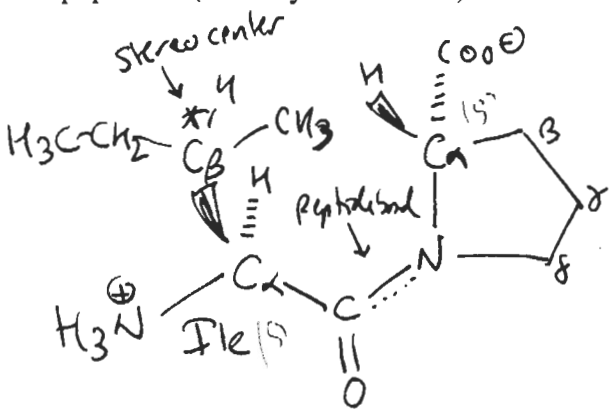
(a; 5 pts) What is the **H-bonding pattern in an α-helix**? You don't need to draw the structure, just specify which atoms are H-bonded to each other. **Why is it important that all of the moieties involved are backbone atoms**, not side-chain atoms?

(+2) 

(+3) Any sequence (except one containing Pro) can form an α-helix - it is independent of side chain identity

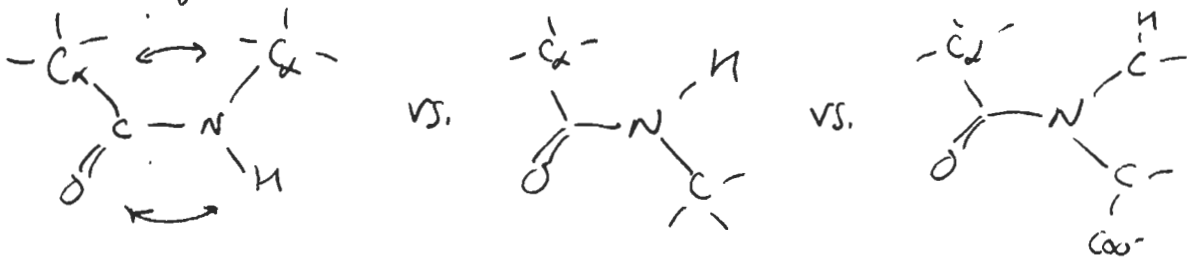
Score for the page_____

(b; 13 pts) Draw **Ile-Pro with a *cis* peptide bond**, in the ionization state observed at pH 7. Include Cα stereochemistry and indicate the Cβ stereocenter on the dipeptide. **Why are X-Pro peptide bonds the only ones that are observed to be either *cis* or *trans***, as opposed to exclusively *trans* for other dipeptides? (X = any amino acid)



(+3) Ile
(+2) ~~+3~~ Pro, (+1) for Cα stereochemistry
~~+3 cis peptide bond~~
(+1) Cβ stereochemistry
(+1) for peptide bond
(+2) for cis peptide bond

(+3) Any other amino acid has a C and an H attached to N – there's a big difference between cis + trans – for Pro it's a C either way:



(c; 5 pts) For each of the five amino acids on the left, **circle the one on the right that is most likely to substitute for it** in a homologous protein.

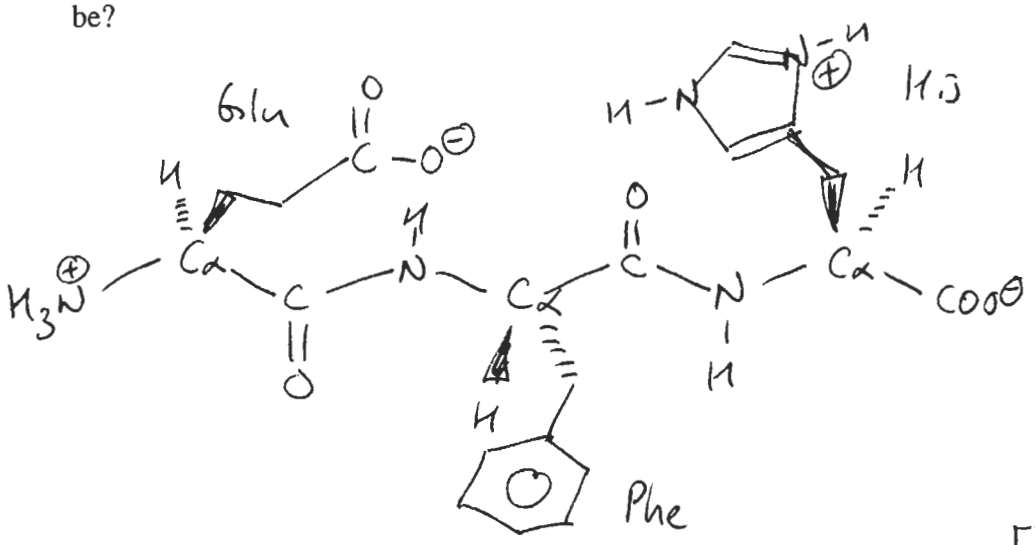
Ile	Leu	(Val)	Asp	Cys
Lys	Glu	Met	(Arg)	Ala
Ser	Tyr	(Thr)	Asn	Ile
Tyr	Gly	(Trp)	Gln	Lys
Glu	Phe	Ser	(Asp)	His

(+1) each, no partial cred. (See Blossum 62 matrix)

Score for the page_____

2. (20 pts) Peptide structure and Henderson-Hasselbach

(a; 10 pts) Draw the **tripeptide EFH** in an extended form (i.e. the way we usually draw peptide sequences), in its predominant ionization state at pH 5. Include the stereochemistry at each Cα. If the tripeptide in this conformation were part of a **secondary structure element**, which one would it be?



+2 Glu (+1 for -COOH)
+2 Phe
+2 His (+1 for)
+2 extended peptide chain
+1 ionization states of termini

[-1 for no stereochem]

→ Looks most like a β strand (or β sheet) +1

(b; 3 pts) It turns out the pKa of the EFH Histidine in this protein is 7.5, rather than whatever it was as an isolated amino acid. (This should not change your answer for the structure.) **Explain why the pKa changed** in this way.

+3

The neighboring ~~Asp~~ Glu-$COO^{\ominus}$ stabilizes the $\oplus$ charge on His H^+ → makes it a weaker acid, raises the pKa.

[+2 for H-bonding between Glu + His, or +3 for proximity of $\ominus$ charge from $COO^{\ominus}$]

Score for the page_____

discussed in part (b)

(c; 7 pts) **At what pH would the average charge on the histidine side chain above be +0.25 (i.e. what pH gives 75% dissociation of $HisH^+$)?**

+2 $pH = pK_a + \log \frac{[His]}{[His\,H^+]}$ $\quad His\,H^+ \rightleftharpoons His + H^+$

we are given that $\frac{[His]}{[His\,H^+] + [His]} = 0.75$ (+2)

so $\frac{([His]/[His\,H^+])}{1 + ([His]/[His\,H^+])} = 0.75$

$[His]/[His\,H^+] = 0.75\,(1 + [His]/[His\,H^+])$

$0.25\,[His]/[His\,H^+] = 0.75$ $\quad [His]/[His\,H^+] = 3$ (+2) (+1)

$pH = 7.5 + \log(3) = 7.97$ (+2) [+1 for arithmetic mistake as long as answer is >7.5]

3. (15 pts) Biomolecules and Molecular Recognition:

(a; 8 pts) Give **two reasons that fat is a denser source of dietary calories** than carbohydrates. When yeast grows fermentatively, it leaves behind an excellent fuel, ethanol. **Why don't the yeast burn the ethanol** to continue growing? **Why do they grow more efficiently under aerobic conditions**?

+2 Fat has more electrons per carbon - it goes from an oxidation state of −2 to +4, whereas $C_n(H_2O)_n$ goes from 0 to +4

+2 Fat packs better into oily globules - carbohydrates carry around all that H_2O, and are solvated by H-bonding to solvent

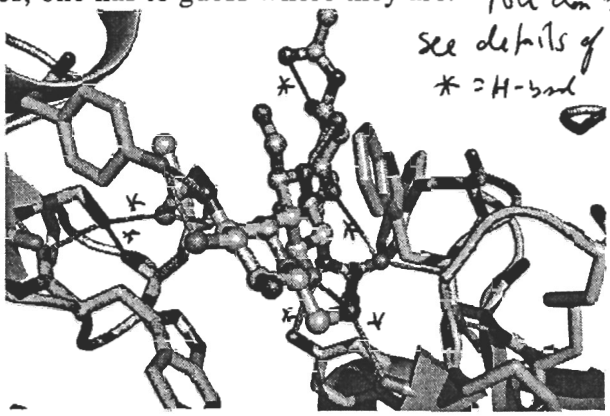
+2 In fermentative growth, the yeast has no terminal electron acceptor - nowhere to dump electrons, can't do combustion

+2 Reduction of O_2 provides large $\ominus$ ΔG.

Score for the page_____

The pictures below show two molecules of a plant lectin protein binding to NAG_3 in the middle; NAG = N-acetylglucosamine, a modified sugar. The dashed lines in the close-up on the right indicate putative hydrogen bonds – since we don't see H in X-ray structures, one has to guess where they are.

You don't need to see details of the image



(b; 7 pts) We claimed that sugars have tremendous information density. **How is the information in sugars encoded and read? Why isn't this encoding suitable for carrying genetic information?**

+2 – Sugars have tremendous variety in size, stereochemistry, functionality, and branching patterns

+2 – Lectins recognize sugars by binding to a pattern of H-bond donors + acceptors, Van der Waals contacts, electrostatics etc.

+3 – This recognition is specific to each lectin/sugar pair, not general like the recognition of DNA by DNA polymerases –

=/= or

+3 No mechanism for copying the genetic information.

4. (22 pts) Tertiary structure and protein folding

(a; 3 pts) We have made the analogy that tertiary and quaternary structure in proteins is held together by Velcro, not nails. **What does this mean in terms of molecular structure and interactions?**

+3 – Proteins are held together by a large # of weak interactions (H-bonds, vdW, e-statics, hydrophobic effect) as opposed to a small number of strong interactions (covalent bonds).

Score for the page_____

(b; 6 pts) How does the Velcro vs. nails analogy help explain a) **cooperative protein (un)folding** and b) **evolution**?

a) (+3) When a protein is unfolded, breaking some weak interactions causes the neighboring interactions to be weakened, so they are also likely to break - net result is an all-or-none unfolding transition.

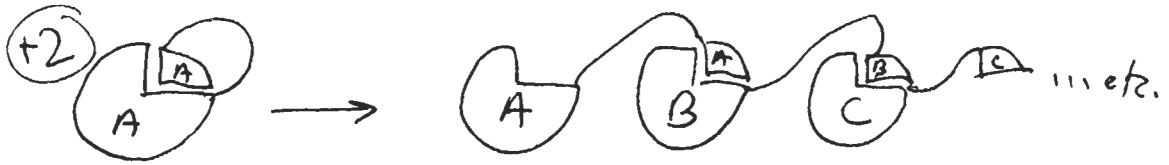
b) (+3) "Weak linkage" allows evolution to gradually tweak structure and function to adapt to new conditions, rather than being rigid or else needing to evolve completely new proteins.

(c; 13 pts) Protein folding and misfolding: A misfolded protein folding intermediate can follow several pathways. **What is ironic about calling the GroEL/GroES chaperone a "foldase"?** The steric zipper is one common failure mode leading from misfolding to protein aggregation. **Name and sketch another mode.** Both modes explain how aggregates can form that contain only one kind of protein, even though the failure modes are generic. **List two different ways in which a protein aggregate might be responsible for disease. What else can happen to a misfolded intermediate (besides chaperone-mediated refolding or irreversible aggregation)?**

(+3) - GroEL/ES actually works as an "unfoldase," giving the target protein a chance to refold correctly on its own.

(+1) - Domain swapping enables specific aggregation:

(+2)



(+2) - The loss of the function of the aggregate can be deleterious

or

(+2) - The aggregate or some intermediate may be toxic, or may provoke an inflammatory or immune response.

(+3) - Misfolded proteins can be recognized by degradative machinery and destroyed by proteolysis

or

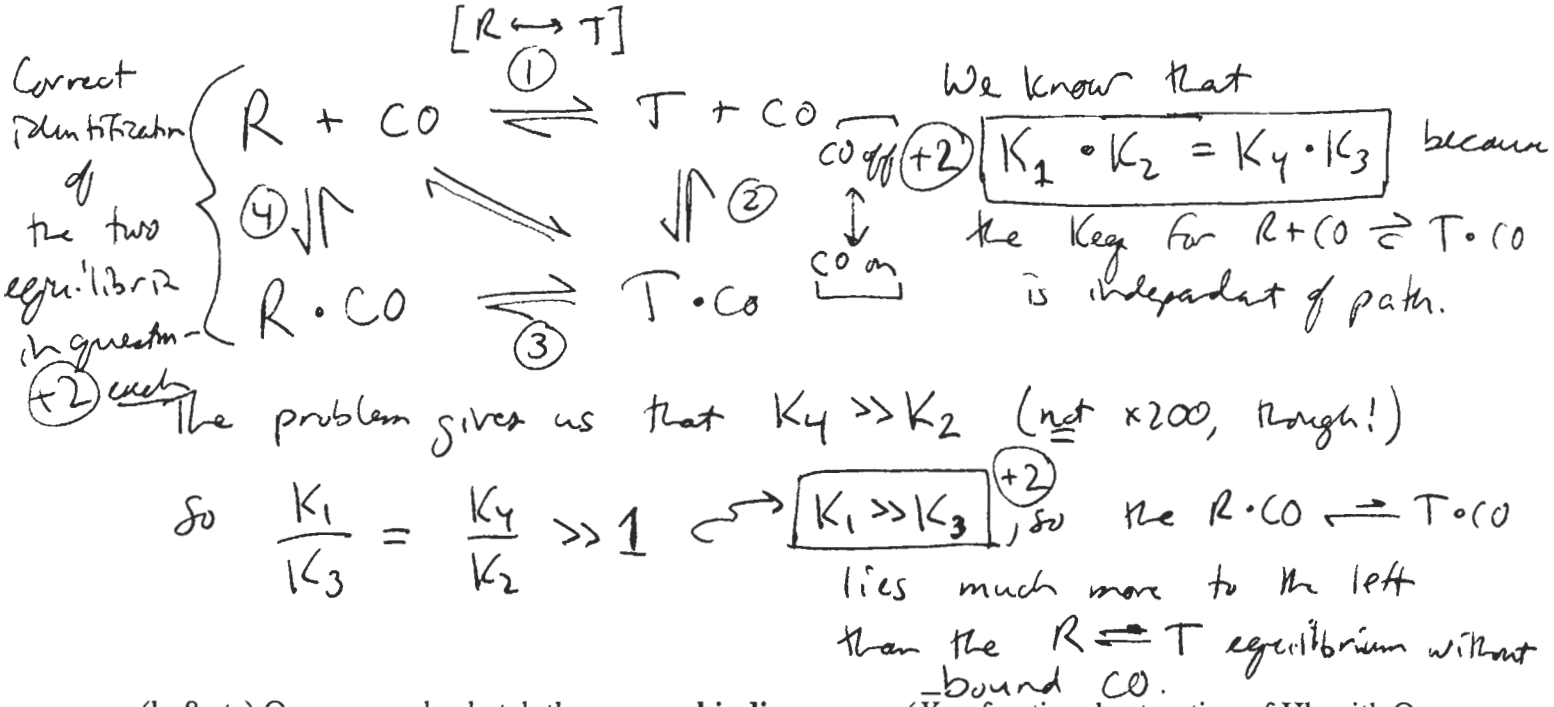
They may spontaneously refold to N.

Score for the page_____

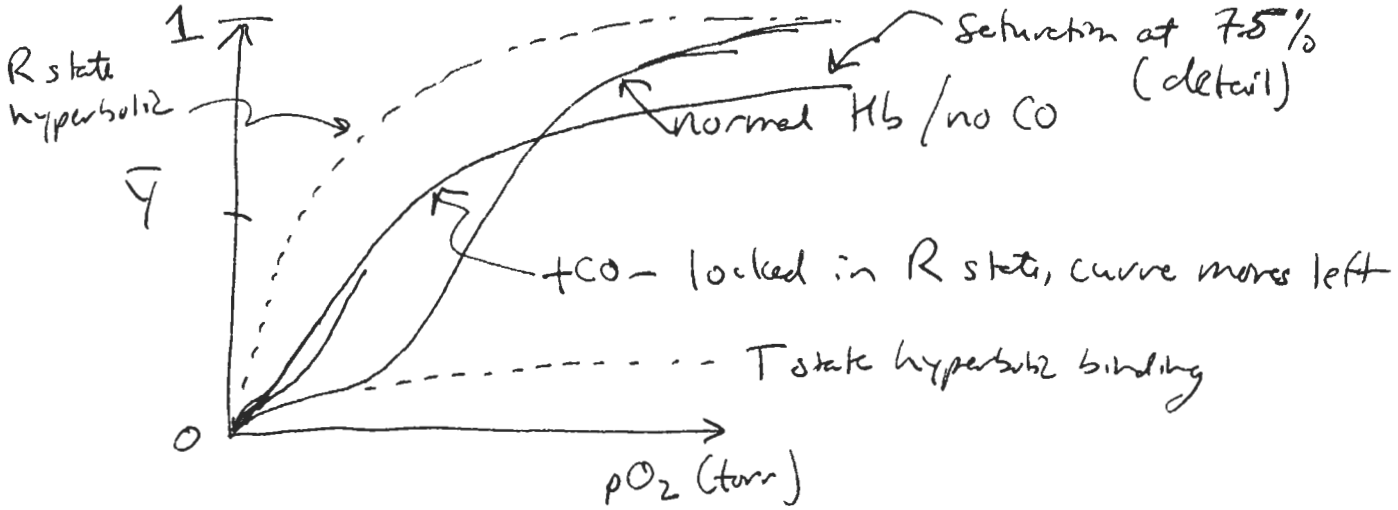
4. (20 pts) Hemoglobin

Carbon monoxide is a poison at least in part because it binds to hemoglobin. Carboxyhemoglobin looks like the O_2-bound state but CO binds with ~200-fold higher affinity than O_2. CO poisoning leads to headache, dizziness, hallucinations, and confusion. Fatal levels CO are much lower than the amount needed to saturate all the O2 binding sites.

(a; 8 pts) CO binds the R state much better than the T state. Sketch and explain the linked equilibria that **demonstrate that CO binding will have the effect of stabilizing the R state.**



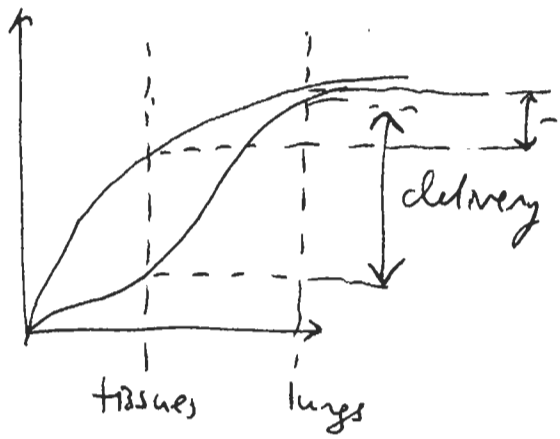
(b; 8 pts) On one graph, sketch the **oxygen binding curves** (Y = fractional saturation of Hb with O_2 vs. pO_2) for (1) **Hb in normal blood** and for (2) **Hb in blood that contains enough CO** to saturate one site per tetramer. There is room on the next page to recopy the graph if you need to.



+2 for idea of graph
+2 for Hb curve
+2 for CO shifting curve left
+1 for more hyperbolic shape for the curve
+1 for all details

Score for the page_____

(c; 3 pts) **How does CO interfere with O_2 delivery?**



(+3) CO prevents Hb from undergoing R → T, prevents dropping off O_2 in tissues

(d; 1 pts) Why do people who live in old houses with faulty furnaces sometimes think that the houses are **haunted** (according to Wikipedia)?

– Incomplete combustion → high [CO] in the air –

(+1) Inhabitants feel lousy and have hallucinations – think the house is haunted.

[This is why they sell CO detectors]

Page	Score
1	/5
2	/18
3	/13
4	/15
5	/10
6	/19
7	/16
8	/4
Total	**/100**

Score for the page_____