Chemistry 240A
Prof. B. Hanson
Spring 2018

Name: KEY

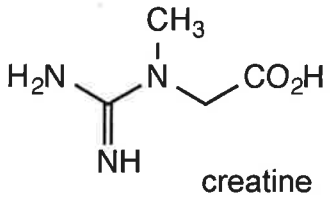
Exam #1

1.	_____	20 pts
2.	_____	20 pts
3.	_____	10 pts
4.	_____	15 pts
5.	_____	15 pts
6.	_____	20 pts
		100 pts

Molecular interactions and chemical transformations are at the heart of biology, and all biological phenomenon that we can analyze today can be traced back to chemical processes: biology is molecular. This insight, on the one hand, makes clear that chemistry may turn out to be the central science in the quest for understanding the molecular basis of life. This is so because the study of interactions between molecules, be they large or small, and the methodology to prepare them and to prepare new compounds with a predetermined set of properties are at the heart of chemistry and constitute the key expertise of chemists. On the other hand, it demands from chemistry to devote a major part of its undisputed powers to addressing the problems unravelled by the research in the biological sciences.

H. Waldman
At the Crossroads of Chemistry and Biology
Bio-organic & Medicinal Chemistry 11 (2003) 3045-3051

1. (20 points). Creatine is an important metabolite that some weight lifters use as a supplement. pK_{a1} is 3.4 and pK_{a2} is 10.6.

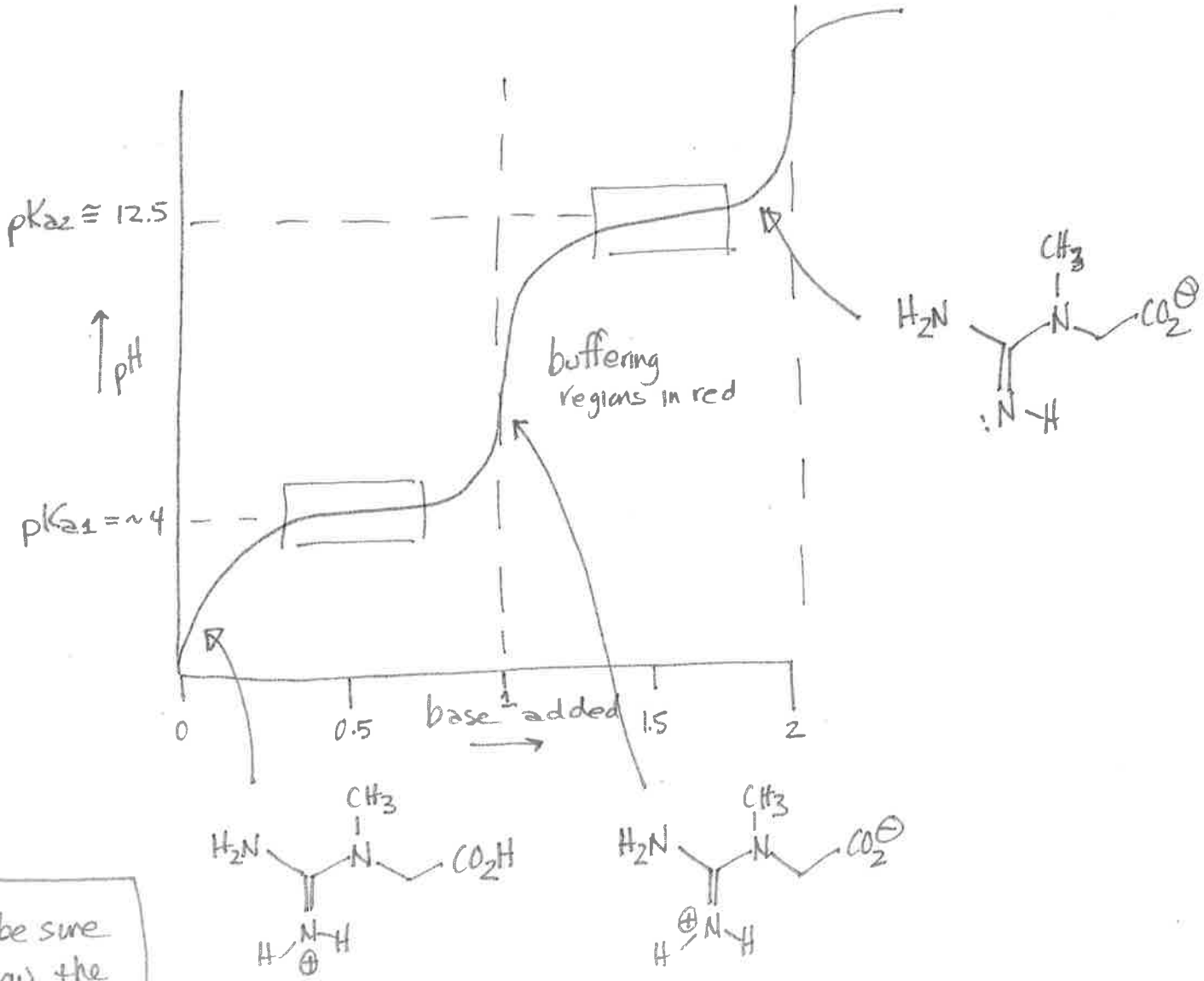


creatine

(a) Sketch and completely label a titration curve for creatine (label each axis; include approximate numeric values). Make your diagram large so you have room for the next items.

(b) If creatine can be a buffer, label your diagram to indicate any buffering region(s).

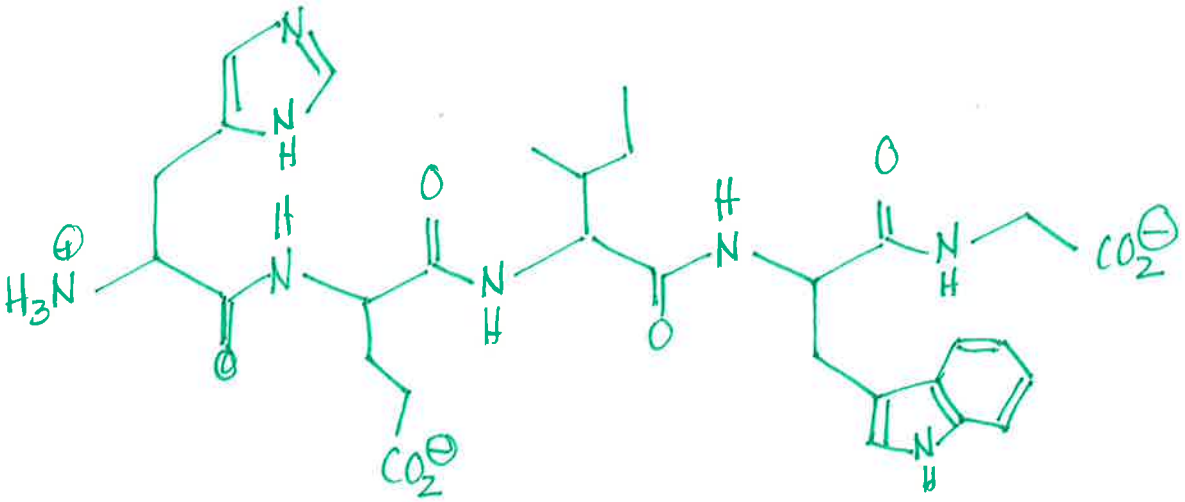
(c) Underneath the titration curve, draw the structure of creatine in its appropriate ionization state at each key point (beginning, end, and at any intermediate point where the ionization state is different/unique).



please be sure you know the acid-base behavior of the guanine group

2. (20 points). Answer the following questions about his-glu-ile-trp-gly

(a) Draw the structure of this peptide as it would appear at pH 7.4. Show all the bonds, but you don't need to show the stereochemistry.



(b) What is the approximate pI of this peptide? Sketch out the relevant equilibria, note the $pK_a's$, and label the species with their charge. No structures needed.

low pH $+2 \overset{pK_a\,3}{\rightleftharpoons} +1 \overset{pK_a\,4}{\rightleftharpoons} 0 \overset{pK_a\,7}{\rightleftharpoons} -1 \overset{pK_a\,10}{\rightleftharpoons} -2$ high pH

$$pI = \frac{4+7}{2} = 5.5$$

(c) What chemical specie or species are present at pH 4.5? No structures needed.

The dominant species is zero-charged, though there is some +1 present

3. (10 points). The features of a Ramachandran plot, and indeed all protein structures, arise from two important concepts from organic chemistry. State these concepts and briefly but carefully describe how they are relevant to protein structure.

(a) Concept 1: Resonance: the amide/peptide bond is planar due to resonance which greatly restricts flexibility

(b) Concept 2: Steric hinderance: collisions of R groups w/ the flat amide grops further restrict flexibility

4. (15 points). This morning I e-mailed you a file which you can save to your desktop and open with Chimera (use *File* → *Open...* then navigate to where you saved the file. Double-clicking probably won't open it).

(a) Give the full identifier of the N-terminus amino acid.

gly 25.A

(b) Are there any disulfide bridges in this structure? If so, give the full identifier of each residue participating in the bridge.

yes cys.118 — cys.99

3 pts each

(c) Are there any cation-π interactions in this protein? If so, give the full identifier of each residue.

Yes, tyr 70.A w/ arg 32.A Another possibility is Phe 121.A + one branch of Arg.37A

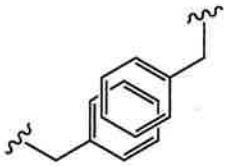
(d) What's going on with arg 37? Why is it "doubled"?

It is disordered in the xtal structure; either position is consistent w the data

(e) Are there any parallel β-sheets in this structure?

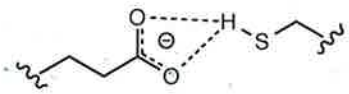
no

5. (15 points). Intermolecular forces are central to understanding protein structure. Shown below are three examples of intermolecular forces typically found in proteins.



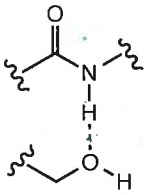
hydrophobic / London forces

3



hydrogen banding

1



hydrogen banding

2

10 pts (a) Give a name for each case, or lacking a name, a descriptive phrase (i.e., what force/attraction is this?)

5 pts (b) Rank these by strength. Use 1 for the strongest interaction. Write the rank under the structure.

this is strongest because the partial charge on the acceptor ($-C^{\delta}(=O)-O^{\ominus}$) is higher than in the other case.

6. (20 points). Imagine that you are working to purify a protein (the target) which is known to have a mass of 120 kDa, and to have a pI of 8.3. It is known that the tissue which contains the target also contains a significant amount of another protein with a pI of 7.5. Let's refer to this second protein as the impurity. There are of course other proteins in the tissue, but we don't have information about them, and they are present at much lower concentrations.

(a) The buffer you choose to use has a pH of 6.8. You decide to use ion-exchange chromatography as your first step. Should you use anion exchange, or cation exchange? State your choice and give a brief, clear explanation.

(target)
At pH 6.8, the main protein is below its pI and will be (+)-ive, while the impurity is in the same situation. You must use cation exchange.

(b) You carry out the ion-exchange experiment that you chose in the previous step. Fractions are collected and labeled A1-A3. Which protein (target or impurity) elutes from the column first? Explain why, succinctly.

The target protein is the farthest from its pI at pH 6.8 so it is the most (+)ive. It will be held most strongly to the resin. Therefore impurity elutes 1st.

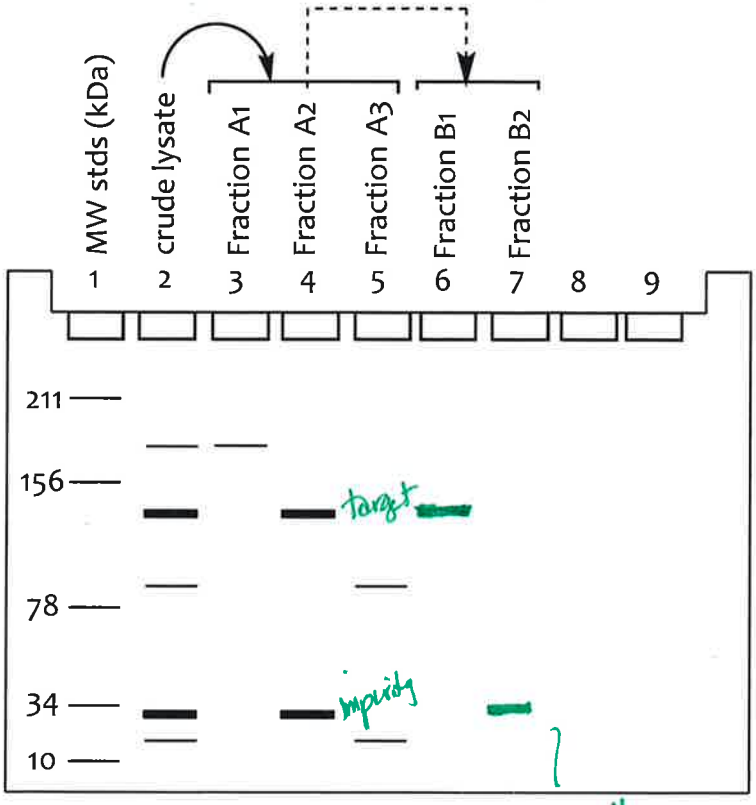
4 questions x 5 pts each

(c) Unfortunately, while the proteins elute at different times, both the protein and the impurity ended up in fraction A2, as shown in the diagram of the gel.

(d) Seeing this result, you decide to do gel filtration on fraction A2, and generate 2 new fractions, B1 and B2. Why is this a good choice given these results?

The MW of the target & impurity are very different.

(e) Now you have great luck - the target and the impurity end up in separate fractions! Fill in lanes 6 and 7 showing the results.



smaller proteins elute later on gel filtration chrom.