C383 Exam 1
Fall 2017

Name Key Seat Number _____

Student ID _____ AI _____

The last page of this exam contains equations, constants, pKa values, and other information you might find useful.
The exam consists of 10 questions worth 100 points plus 5 bonus points on a total of 11 pages. It will be scored out of 100 points.

1-15 _____/30 multiple choice

16-30 _____/30 fill in the blank

31 _____/10

32 _____/10

33. _____/10

34. _____/10

Bonus _____/5

Total:

Regrading: All requests for regrades must be submitted in writing within 48 hours of the return of the exam. You must explicitly state what has been misgraded and why it is an error. The entire exam will be regraded, which could result in points being added or deducted overall.

Section 1: Multiple Choice. 15 questions, 2 points each.

1. B Which of the following bonds serves as a hydrogen bond donor in nucleotide base pairing??

A) C=O
B) N–H
C) O–H
D) P=O
E) None of the above.

2. B What is the $[A^-]/[HA]$ ratio of an acid with pKa = 7.5 when it is in a solution at pH 6.5?

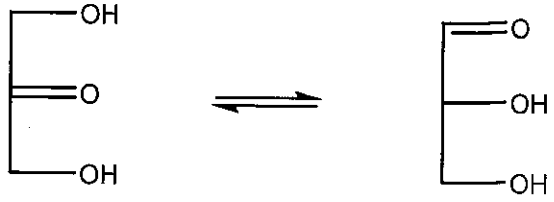
A) 1:1
B) 1:10
C) 10:1
D) 2:1
E) None of the above.

3. C The net charge on tyrosine at pH 7.4 is approximately

A) +2
B) +1
C) 0
D) -1
E) -2

4. A Which of the following amino acid residues would be more likely found in an irregular loop or a turn than in an alpha helix or beta sheet?
A) proline
B) serine
C) phenylalanine
D) lysine
E) glutamine

5. E What class of enzyme would catalzye the following reaction?



A) oxidoreductase
B) transferase
C) lyase
D) ligase
E) isomerase

6. B Which of the following techniques would be best for separating samples of the following peptides: ADTEDDVSWEDQIL and KRSTWCWRRHMPK?

A) Edman degradation
B) Ion exchange chromatography
C) SDS-PAGE
D) homogenization
E) Gel filtration chromatography

7. E A spontaneous reaction reaches equilibrium when

A) its ΔG becomes negative.
B) its ΔG becomes positive.
C) its $\Delta G^{0'}$ becomes zero.
D) its $\Delta G^{0'}$ becomes negative.
E). none of the above

8. A You determine that a protein you are studying is made up of two distinct polypetides. The two polypeptides might be covalently linked through a _____ bond.

A) disulfide
B) peptide
C) hydrogen
D) ester
E) More than one of the above

9. A Which of these amino acids would not be capable of being involved in a salt bridge interaction at pH 5?

A) Gln
B) K
C) His
D) R
E) None of the above

10. E The _____ model of enzyme substrate binding takes into account the dynamic nature of the enzyme-substrate interaction.

A) transition state analog
B) lock and key
C) rate enhancement
D) active site
E) induced fit

Accept any

11. _____ You performed a Meselson and Stahl type experiment in which you had bacteria starting with all ^{15}N DNA, and then fed all ^{14}N precursors. If you grew the bacteria for one generation and then isolated the DNA of the daughter cells, what would you expect for the composition of the ~~two bands~~? (Assume semi-conservative replication.)

A) One DNA band that is all ^{14}N
B) One DNA band that is all ^{15}N.
C) One DNA band with mass intermediate between ^{14}N and ^{15}N.
D) Two bands, one all ^{14}N and one all ^{15}N.
E) Two bands, one all ^{14}N and one with mass intermediate between ^{14}N and ^{15}N.

12. C The specific activity of a purified sample of enzyme can be determined if the total activity and the _____ are known.

A) yield
B) specificity constant
C) total protein
D) purification level
E) assay

13. C The isoelectric point of the lysine-aspartate dipeptide is about

A) 2
B) 4
C) 6
D) 8
E) 10

14. B If the composition of a sample of double stranded DNA is found to be 20% A, then the composition of G would be

A) 20%
B) 30%
C) 40%
D) 50%
E) Cannot be determined from these data.

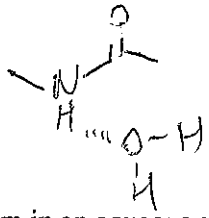
15. D Which of the following statements is not true concerning the protein collagen?

A) Collagen is a fibrous protein.
B) Mutations in collagen can have a large impact on connective tissue.
C) Collagen contains glycine at every third residue.
D) Collagen has a coiled-coil structure.
E) Scurvy is caused by the inability to make hydroxyproline necessary for collagen production.

Section 2: Fill in the blank. 15 questions 2 points each

16. A phosphorylated ribose with a nucleobase attached is called a nucleotide.

17. Draw a hydrogen bond between water and an amide in which water is the H-bond acceptor.



18. The concentration of hydronium in an aqueous solution at pH 9 is 1×10^{-5} M.

19. Two hydrophobic, aromatic amino acids are Phe and Trp.

20. The hydrophobic effect explains why it is thermodynamically favorable for a phenylalanine sidechain to become buried in the center of a globular protein.

21. An essential amino acid must be eaten in the diet because it cannot be synthesized biochemically.

22. Regular secondary structures stabilize protein folding primarily by maintaining hydrogen bonding on the interior of globular proteins.

23. Alzheimers is a disease correlated with protein misfolding. (Others accepted)

24. An imidazole buffer would be effective in the pH range of 6.0 to 8.0.

25. DNA is a long, unbranched polymers consisting of monomers joined together by the phosphodiester linkage.

26. The one letter abbreviation for the amino acid glutamine is Q.

27. The difference in structure between RNA and DNA is mainly caused by the presence of 2' hydroxyl in RNA, which allows for more options for hydrogen bonding.

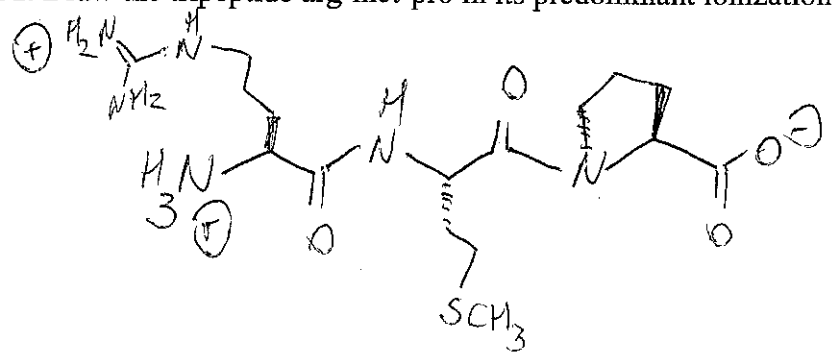
28. SDS-PAGE is a technique that can be used to separate proteins based on their size.

29. The tightly bound coenzyme of a holoenzyme is called a prosthetic group.

30. Enzymes accelerate the attainment of equilibrium, but do not shift its position, which is a function of the difference in free energy.

Section 3. Problems. 4 questions 10 points each.

31. Draw the tripeptide arg-met-pro in its predominant ionization state at pH 7.

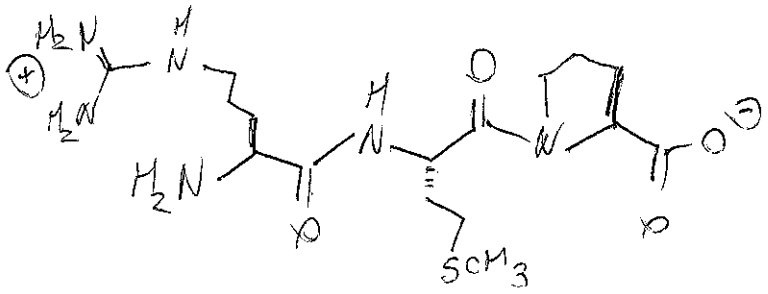


+1 per side chain
+1 backbone
+1 charge

What is the isoelectric point of this tripeptide? Show all calculations, and then draw the tripeptide in its predominant ionization state at its isoelectronic point.

$$pI = \frac{12.5 + 8.0}{2} = 10.2$$

+3



How would the tripeptide pro-met-arg be structurally different than the tripeptide you drew above? Would you expect to be able to separate these two dipeptides by ion exchange chromatography?

+2

* Differ in N-terminus and C-terminus end.
* Difficult to separate

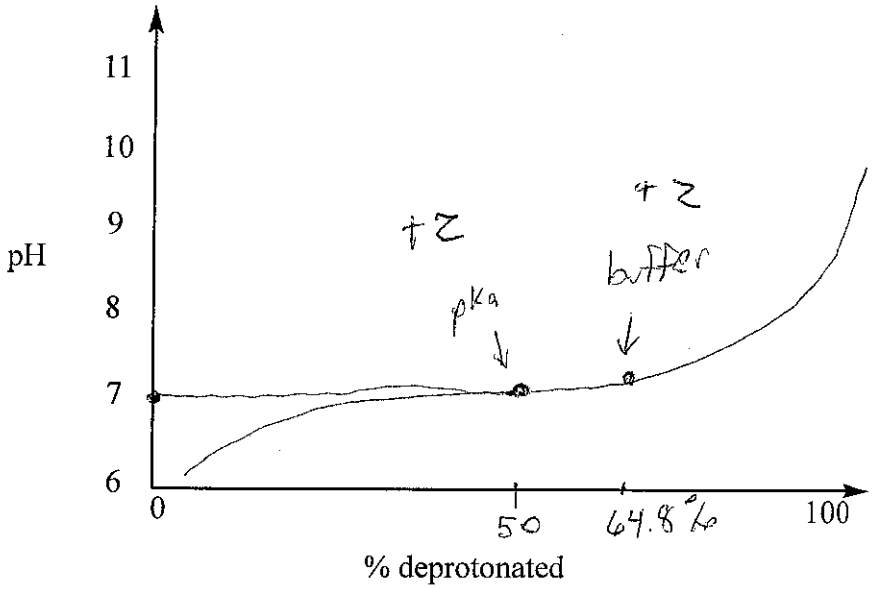
32. To a solution containing 50.0 mmol of the protonated form of a buffer called ACES (pKa = 6.90) was added 32.4 mmol of sodium hydroxide.

A. What is the pH of this solution?

$$pH = pK_a + \log \frac{A^-}{HA}$$

$$+3 \quad = 6.9 + \log \frac{32.4}{17.6} = \boxed{7.2}$$

B. Draw a titration curve for ACES, and indicate where on the curve this buffer is located.



C. If the pH of the solution were changed to 6.7, what percent of the buffer molecules would be in their conjugate base form?

$$pH = pK_a + \log \frac{A^-}{HA}$$

$$+3 \quad 6.7 = 6.9 + \log \frac{A^-}{HA}$$

$$\frac{A^-}{HA} = .63$$

$$\%A^- = \frac{.63}{1.63} = 39\%$$

33. Consider the following reaction:

Glucose-1-phosphate $\rightleftharpoons$ Glucose-6-phosphate

These compounds were placed in a beaker and the reaction was allowed to proceed to equilibrium, at which time [glucose-1-phosphate] = 0.010 M and [Glucose-6-phosphate] = 0.19 M.

A. What is $\Delta G^{0'}$ for the reaction in this beaker?

+4

$$\Delta G' = \Delta G^{0'} + RT \ln \frac{[G\text{-}6\text{-}P]}{[G\text{-}1\text{-}P]}$$

$$0 = \Delta G^{0'} + 8.314 \frac{J}{mol\,K} (298\,K) \ln \frac{0.19\,M}{0.010\,M}$$

$$\Delta G^{0'} = -7.3 \frac{kJ}{mol}$$

B. What ΔG for the reaction in this beaker?

+3

$\Delta G'$ = Zero for any rxn at equilibrium

C. What is K_{eq} for the reaction in this beaker?

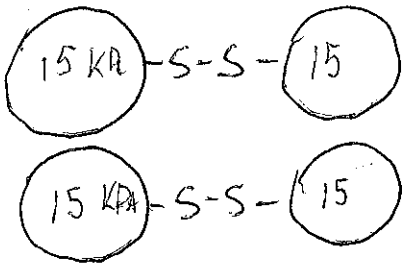
+3

$$K_{eq} = \frac{0.19\,M}{0.010\,M} = 19$$

34. Protein Purification

A. A protein was purified to homogeneity. Determination of the mass by gel-filtration chromatography yields 60 kDa. Chromatography in the presence of urea yields a 30 kDa species, When the chromatography is repeated in the presence of urea and β mercaptoethanol, a single molecular species of 15 kDa results. Draw a representation of the molecule.

+5



Dimer of dimers.

Each dimer is connected via a disulfide bond

B. The octapeptide AVGWRVKS was digested with the enzyme trypsin. Would ion exchange or size exclusion chromatography be more appropriate for separating it products? Explain.

AVGWR | VK | S

↓ trypsin

same charge

AVGWR

VK

S

different by size:

Size exclusion would be better

+1

+2

+2

Bonus: A protein purification scheme was conducted as outlined below, and the purification data is summarized in the table.

Procedure	Total protein (mg)	Total activity (units)	Specific activity (units/mg)	Purification level	Yield (%)
Crude extract	20,000	4,000,000	200	1	100
Salt precipitation	5,000	3,000,000	600	3	75
Ion exchange chromatography	1500	1,000,000	667	3.3	25
Gel-filtration chromatography	500	750,000	1500	7.5	19
Affinity Chromatography	45	675,000	15,000	75	17

Which of these steps was most effective in the purification scheme? Explain how you came to this answer. **If you have a reasonable guess, go for it! You may receive up to 5 points bonus on this problem with possibility of partial credit, but <u>answers that show no understanding will receive a -1 penalty</u>. Better to leave it blank than to make a wild guess.**

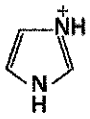
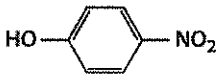
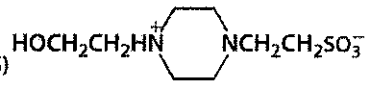
Affinity chromatography has a 10x increase in purification level. +5

Useful Information:

$$\Delta G_{reaction} = \Delta G^{\circ\prime}_{reaction} + RT \ln \frac{[products]}{[reactants]}$$

$\Delta G^{0'} = -RT \ln K_{eq}$ R = 8.314 J/mol.K $pH = pK_a + \log [A^-]/[HA]$

[TABLE 2.4] pK Values of Some Acids

Name	Formula [a]	pK
Trifluoroacetic acid	CF_3COOH	0.18
Phosphoric acid	H_3PO_4	2.15[b]
Formic acid	$HCOOH$	3.75
Succinic acid	$HOOCCH_2CH_2COOH$	4.21[b]
Acetic acid	CH_3COOH	4.76
Succinate	$HOOCCH_2CH_2COO^-$	5.64[c]
Thiophenol	C_6H_5SH	6.60
Phosphate	$H_2PO_4^-$	6.82[c]
N-(2-acetamido)-2-amino-ethanesulfonic acid (ACES)	$H_2NCOCH_2\overset{+}{N}H_2CH_2CH_2SO_3^-$	6.90
Imidazole		7.00
p-Nitrophenol		7.24
N-2-hydroxyethylpiperazine-N'-2-ethanesulfonic acid (HEPES)		7.55
Glycinamide	$^+H_3NCH_2CONH_2$	8.20
Tris(hydroxymethyl)-aminomethane (Tris)	$(HOCH_2)_3C\overset{+}{N}H_3$	8.30
Boric acid	H_3BO_3	9.24
Ammonium ion	NH_4^+ 9.25	
Phenol	C_6H_5OH	9.90
Methylammonium ion	$CH_3NH_3^+$	10.60
Phosphate	HPO_4^{2-}	12.38[d]

Amino acid pKa values

C-terminal	3.1
N-terminal	8.0
Aspartate, glutamate	4.1
Histidine	6.1
Cysteine	8.3
Tyrosine	10.9
Lysine	10.8
Arginine	12.5

Table 5.3 Specific cleavage of polypeptides

Reagent	Cleavage site
Chemical cleavage	
Cyanogen bromide	Carboxyl side of methionine residues
O-Iodosobenzoate	Carboxyl side of tryptophan residues
Hydroxylamine	Asparagine-glycine bonds
2-Nitro-5-thiocyanobenzoate	Amino side of cysteine residues
Enzymatic cleavage	
Trypsin	Carboxyl side of lysine and arginine residues
Clostripain	Carboxyl side of arginine residues
Staphylococcal protease	Carboxyl side of aspartate and glutamate residues (glutamate only under certain conditions)
Thrombin	Carboxyl side of arginine
Chymotrypsin	Carboxyl side of tyrosine, tryptophan, phenylalanine, leucine, and methionine
Carboxypeptidase A	Amino side of carboxyl-terminal amino acid (not arginine, lysine, or proline)

Table 5.3
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