

October 12, 2005
Fall 2005
Isom

EXAM 2
Biochemistry I / CHEM 4320

Name: _____

1) a) Assemble the full peptide (pep A) sequence from the following fragments (5pts)

CNBr digestion

Pro-Trp-Cys-Arg-Met
Cys-Phe-Met
Cys-Ile-Tyr-Met

Chymotrypsin digestion

Met-Pro-Trp
Met-Cys-Phe
Cys-Ile-Tyr
Cys-Arg-Met

b) Write the sequence you determined in (a) below and draw the location of all disulfide bonds. Chymotrypsin digestion of the intact peptide followed by acid hydrolysis produced the following fragments. (6 pts)

(pep a)

(pep b) Val-Cys-Trp-Val-Cys-Phe-Gly-Cys-Lys

(Lys, Gly, Trp, Val, Cys₂)

(Arg, Val, Cys₂, Phe, Met)

(Met, Pro, Trp)

(Phe, Tyr, Ile, Met, Cys₂)

c) Draw the C-terminal CNBr fragment as expected at pH 7.4. Circle a peptide bond. (13 pts)

2) A group of people **having the mutation present in Sickle Cell anemia** were found not to produce Hb polymers and so have no symptoms of the disease. What **specific** changes in primary structure could account for this observation? Explain the specific changes and your reason for choosing them. (6 pts)

3) While investigating protein folding, a researcher notices that one type of secondary structure tends to form at an earlier stage of protein folding than the other. What do you think she concluded and why? (6pts)

4) Based on what you know about chaperones, which model concerning the prevention of kinetically stable intermediates seems the most correct and why? (6 pts)

5) Explain the result of the following mutations on protein folding and GroEL/GroES conformation and efficiency (assuming all subunits fold and assemble normally) (12 pts)

a) 50% increase in hydrophobic residues in GroEL; (b) a large increase in disulfide bonds within subunits; (c) 50% increase in the expression of GroEL

6) A researcher isolates the following peptides from an organism that is found to be susceptible to a prion-like malady involving protein aggregation.

(A) Arg-His-Lys-His-Phe-Ala-Val-Trp-Leu-His-His-Arg-His (B) Glu-Asp-Glu-Asp-Phe-Tyr-Phe-Trp-Phe-Glu-Glu-Asp-Asp

a) One protein forms a fairly stable α -helix in the cytoplasm is found to lose this structure stability inside lysosomes and to produce plaques containing an insoluble pentapeptide fragment after digestion by a trypsin-like protease. No such fragment is produced when the protein is incubated with this protease in the cytoplasm. Which protein is responsible for these plaques and explain the researcher's observations. (10 pts)

b) The other protein tends to aggregate as a whole in the lysosome. Analysis of the plaques indicates that the aggregated protein has an α -helical structure that is not present in its cytoplasmic form. Explain these observations. (10 pts)

7) What primary and secondary structure would you expect to be present in the PrP^C octapeptide repeats? Explain. (6pts)

8) You must decide to take one of the following who are heterozygous for the following Hb mutations on a hike up a mountain with you. Assume none of the mutations are fatal. Who would you pick and why? (12 pts)
(a) David (His E7 to Leu) (b) Susan (Tyr 42 to Ser) (c) Rachael (Asn 102 to His)

9) A researcher isolates plaques from a prion victim and wants to conclusively decide which barriers were present and overcome in the victim. She isolates the protein and finds that the amino acid sequences of the aggregated proteins are all identical, but does notice some difference in glycosylation among the proteins. Which barriers can she conclusively say are present in this case and which can she rule out? Briefly explain. (8 pts)
