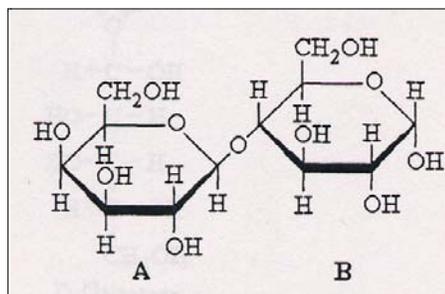


BIOCHEMISTRY 300 CHRISTMAS EXAM, 2001

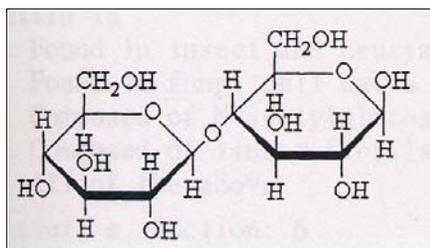
Marks

I. Multiple Choice Questions (1 mark each)

1. The structure of a disaccharide is shown below. Which statement applies?
- Both rings A and B are in equilibrium with an open chain form.
 - Only ring A is in equilibrium with an open chain form.
 - Only ring B is in equilibrium with an open chain form.
 - Neither ring is in equilibrium with an open chain form.



2. What is the name of the disaccharide shown below that is formed by joining two monomers of D-glucose?
- β -D-glucopyranosyl-(1 \rightarrow 4)- β -D-glucopyranose
 - α -D-glucopyranosyl-(1 \rightarrow 4)- α -D-glucopyranose
 - β -D-glucofuranosyl-(1 \rightarrow 4)- β -D-glucofuranose
 - α -D-glucopyranosyl-(1 \rightarrow 4)- β -D-glucopyranose



3. Which is not a similarity between active transport proteins and enzymes?
- Both undergo conformational changes upon binding a substrate.
 - Both are susceptible to inhibition.
 - Both cause chemical modification to the substrate.
 - Both can reach a saturation limit.
4. What is the role of cholesterol in animal cell membranes?
- Blocks the association of the fatty acyl chains of phospholipids at high temperature.
 - Aids in the transport of small hydrophobic molecules across the membrane.
 - Is a receptor site for hormones on the surface of membranes.
 - Broadens the temperature range of optimum membrane fluidity.
5. Which statement is false about lipid-anchored membrane proteins?
- They are found only on the outer leaflet of the plasma membrane.
 - Like integral membrane proteins, they are permanently associated with the membrane.

- c) The covalent link to the membrane involves the phosphate group of the lipid anchor.
 d) The protein portion that can be removed by treatment with phospholipase most resembles an integral membrane protein.
6. Which does not apply to the diffusion of O₂, CO₂ and small hydrophobic molecules across a membrane?
- a) Diffusion is driven by the concentration gradient across the membrane.
 b) The diffusion is spontaneous and there is a decrease in free energy as diffusion occurs.
 c) The transport is saturatable.
 d) Membrane proteins are not needed for the diffusion process.
7. The membrane transport protein Na⁺-K⁺ ATPase carries both Na⁺ and K⁺ ions across the plasma membrane. Typically the concentration of K⁺ inside cells is about 30 times higher inside the cell than outside. The concentration of Na⁺ is about 20 times less inside the cell than outside. Based on this information, which statement below is false?
- a) The proper functioning of Na⁺ - K⁺ ATPase could serve as an energy source for secondary active transport proteins.
 b) It is a symport.
 c) Transport of Na⁺ and K⁺ must be coupled to an exergonic reaction.
 d) Na⁺-K⁺ ATPase likely undergoes conformational changes during transport.
8. Ethanolamine, serine and choline can be cleaved from glycerophospholipids by treatment with
- a) phospholipase A₁
 b) phospholipase A₂
 c) phospholipase B
 d) phospholipase C
 e) phospholipase D
9. The hydrolysis of ATP to ADP has a $\Delta G^{\circ'}$ of -30 kJ/mol. If in an *E. coli* cell the concentrations of ATP, ADP and inorganic phosphate are 7.90 mM, 1.04 mM and 7.9 mM, respectively, which statement is true about the hydrolysis of ATP in the cell? (Assume a temperature of 298 K)
- a) Hydrolysis can proceed spontaneously.
 b) The hydrolysis is at equilibrium.
 c) The formation of ATP from ADP is occurring more rapidly than the hydrolysis.
 d) The hydrolysis is being allosterically controlled.
10. The conversion of glucose-1-phosphate to glucose-6-phosphate by the enzyme phosphoglucomutase has a $\Delta G^{\circ'}$ of -7.6 kJ.mol. Calculate the equilibrium constant for this reaction at 298 K and a pH of 7.
- a) 0.003
 b) 0.047
 c) 1.00
 d) 21
11. A compound with a greater free energy of hydrolysis than ATP is

- a) glucose 1-phosphate.
 - b) any phosphoester.
 - c) phosphoenolpyruvate.
 - d) acetyl CoA.
 - e) all of the above
12. ATP is thermodynamically suited as a carrier of phosphoryl groups in animal cells because
- a) it is stable under cell conditions.
 - b) it is not hydrolyzed in cells without enzyme action.
 - c) it is intermediate in group-transfer potential.
 - d) it can be produced from phosphocreatine
 - e) all of the above
13. Yeast will normally convert pyruvate to ethanol. Why is this better for the yeast than conversion to lactate?
- a) Conversion to ethanol releases more NAD⁺ per mole than the conversion to lactate.
 - b) The carbon atoms are more oxidized in ethanol than in lactate.
 - c) Ethanol is neutral, but lactate production is accompanied by a sharp decrease in pH.
 - d) Ethanol production is not better. Yeast normally produces ethanol and lactate in equimolar amounts.
14. Which is not among the possible fates of pyruvate after glycolysis?
- a) conversion to lactate
 - b) further reduction by the citric acid cycle
 - c) conversion to ethanol
 - d) used in the biosynthesis of alanine
15. Hamsters love to run on exercise wheels. Prolonged running at a high rate of speed requires ATP. Could a hamster with a defective gene for the enzyme lactate dehydrogenase meet the extra ATP demand for prolonged, fast wheel-running by maintaining a high rate of glycolysis? Why or why not?
- a) No, not enough NAD⁺ can be regenerated for glycolysis to continue at a high rate.
 - b) No, the defective gene will cause a rapid decline in pH in the muscles used for running.
 - c) Yes, the defective enzyme has no effect on the glycolytic pathway.
 - d) Yes, the enzyme alcohol dehydrogenase will supply the needed NAD⁺ if the lactose dehydrogenase cannot.
16. Seven of the ten reactions in the glycolytic pathway have free energy values close to zero. What does this tell us about those reactions?
- a) They are near equilibrium reactions.
 - b) They are not control points for pathway regulation.
 - c) They are reversible reactions.
 - d) All of the above
 - e) None of the above
17. The overall ΔG for glycolysis is -72 kJ/mol in erythrocytes. Which statement below is true?

- a) The value of ΔG° is also -72 kJ/mol since the cytosol pH is close to 7.
- b) The free energy of glycolysis is found as the sum of the standard free energy changes for the individual pathway reactions.
- c) The negative sign of ΔG shows that this pathway will proceed toward product (pyruvate) under normal cellular conditions.
- d) All of the above
18. There are four enzymes that can catalyze the first step of glycolysis. They are hexokinases I, II and III and glucokinase (hexokinase IV). Hexokinases I, II and III have K_M values near 0.1 mM. Glucokinase has a K_M of 2-5 mM. If after a heavy meal the blood glucose level rises to 6 mM, which statement will be true?
- a) At this high blood glucose level, all four hexokinases are saturated with substrate.
- b) Hexokinases I, II and III are catalyzing at their maximum rate, but glucokinase can still respond to increases in blood glucose levels.
- c) None of the enzymes is saturated. All of them help to increase the rate of glycolysis.
- d) The flux through glucokinase will be extremely low. The only significant catalysis is done by hexokinases I, II and III.
19. ATP is a cosubstrate of the enzyme PFK-1. In most species ATP is also an inhibitor of PFK-1 at higher concentrations. Which statement below would best explain this?
- a) PFK-1 must be phosphorylated by ATP in the active site and the phosphorylated PFK-1 must be the less active form.
- b) There must be another cofactor interacting with ATP at high concentrations to achieve inhibition of PFK-1.
- c) ATP actually activates the reverse of the reaction preceding the PFK-1 step in the pathway. It likely has no direct effect on PFK-1.
- d) There are two sites on PFK-1 that bind ATP. One is the active site; the other is the regulatory site where inhibition occurs.
20. More than one step in the glycolytic pathway is subject to regulation. It might seem most efficient to regulate only the first step of a pathway to avoid buildup of intermediates and to conserve materials and energy. Why is the first step of glycolysis not the only regulated step?
- a) Some sugars can enter the glycolytic pathway beyond the first step. If steps other than step one were not regulated, the breakdown of these sugars would be essentially uncontrolled.
- b) Having more than one regulated step in the pathway allows for feedback inhibition.
- c) Control of a single step in a reaction pathway is difficult because the concentrations of enzymes in cells are very low. It is easier to control more than one enzyme than just one.
- d) All the ATP in a cell would be depleted very quickly if only the first step of glycolysis were regulated.

II. Questions to be answered on the paper

Marks

- 5 1. a) What residues constitute the catalytic triad of chymotrypsin? What type of catalysis does each mediate?
- b) What groups are found in the oxyanion hole? What type of catalysis do they mediate?

- c) Explain why site-directed mutagenesis of aspartate to asparagine in the active site of chymotrypsin decreases catalytic activity 10,000-fold.
- 5 2. A researcher plans to make a synthetic pentapeptide containing one acidic, one basic, one aromatic and one long chain aliphatic residue, as well as a residue which would allow formation of a disulphide bond. Using the three letter nomenclature, provide an example of a peptide primary structure which would meet this requirement.
3. In a few brief words, answer the following questions about the peptide you have chosen above:
- 2 (a) What would be the estimated net charge on this peptide at pH 4?
- 2 (b) Would it bind to a cation exchange column at pH 11? Why?
- 1 (c) How would you accurately determine its molecular weight experimentally?
- 2 (d) What would you have to do before determining the peptide amino acid composition? How?
- 2 (e) Name two methods which you might employ to verify the amino acid sequence.
- 4 (f) A disulphide bridge is a problem in sequence analysis. How would you remove it? Draw the complete structure of the modified amino acid only.
- 2 4. Name two methods by which this researcher could detect the elution of this peptide from a chromatographic column.
- 10 5. a) Describe the sequence of biochemical events that occurs between the release of epinephrine into the bloodstream and the activation of the enzyme glycogen phosphorylase.
b) Signals resulting from increases in hormone levels are eventually terminated. The response only continues for a limited time. Discuss three different mechanisms for signal termination, using specific systems as examples.
- 10 6. a) Describe the role of G proteins in olfactory sensory transduction.
b) Based on what you know about G proteins, describe the molecular basis for the toxic effects of pertussis toxin and cholera toxin.
- 5 7. Glucose can be oxidized directly to CO_2 and H_2O in a single step. Why does a biological system use so many steps?
- 10 8. Draw the complete structure of NAD^+ , showing the reduced and oxidized forms of the ring. Identify the types of bonds that join the various components of the molecule together. What properties of the molecule account for its central role as a cofactor in biological redox reactions?
- 10 9. If the concentration of Ca^{2+} inside mammalian cells is about $1 \mu\text{M}$, and the concentration of the ion in blood plasma is about 1mM , for cells with a transmembrane potential of -0.08V (inside negative relative to outside), what is the free energy change for transporting 1mol of Ca^{2+} out of the cells at 37°C ?
- 10 10. Show how an enzyme like PFK-1 can be controlled by both covalent and allosteric mechanisms.

END

BIOCHEMISTRY 300 CHRISTMAS EXAM, 2002

Marks

- 5 1. Describe high performance liquid chromatography as it pertains to proteins – also state

- 5 2. Describe the basic principle of mass spectrometry and why laser assisted desorption/ionization technology was an important technical advance for protein biochemists. For simplicity confine your answer to the analysis of a whole intact protein. You do not need to provide any mathematical equations.
- 5 3. Draw 4 amino acids in the structure of an alpha helix. You do not need to draw the R groups, simply put an "R" where the groups belong. Draw the carboxy terminal end at the bottom of the helix. Explain in words all the important structural features you are attempting to draw. Make sure you draw the H bond(s) that stabilize the structure.
- 5 4. Describe the Bohr effect.
- 10 5. You measure the initial rate of an enzyme reaction as a function of substrate concentration in the presence and absence of an inhibitor and obtain the following data:

[S] (M)	v ($\mu\text{mol min}^{-1}$) - Inhibitor	v ($\mu\text{mol min}^{-1}$) + Inhibitor
0.0001	33	17
0.0002	50	29
0.0005	71	50
0.001	83	67
0.002	91	80
0.005	96	91
0.01	98	95
0.02	99	98
0.05	100	99
0.1	100	100

- (a) What is the V_{max} in the absence of inhibitor?
 (b) What is the K_m in the absence of inhibitor?
 (c) When $[S] = 0.0004$ M, what will v be in the absence of inhibitor?
 (d) When $[S] = 0.0004$ M, what will v be in the presence of inhibitor?
 (e) If no inhibitor is present, what fraction of the enzyme is bound to substrate at $[S] = 0.0004$ M?
- 6 6. Illustrate and explain why the conversion of lactate to pyruvate may be favoured in liver, while the reverse reaction may be favoured in muscle.
- 8 7. Draw the complete structure of any specific molecular species of phosphatidylcholine. Indicate where the molecule is cut by phospholipases C and D.
- 8 8. Identify the major components and the biological roles of the following:
 Chitin
 Waxes
 Hyaluronate
 Gangliosides
- 8 9. Diagram and explain the free energy changes that might occur if a molecule like glucose were to cross from one side of a bilayer to the other by simple diffusion. Show how and why the free energy changes might be different when glucose crosses the bilayer via glucose permease (the glucose transporter in cells like erythrocytes).

- 8 10. What are isozymes? Use three different enzymes and the reactions they catalyze to illustrate the importance of isozymes in biological systems.
- 8 11. With the aid of diagrams, illustrate the dependence of the velocity of the reaction catalyzed by phosphofructokinase I on the concentration of fructose-6-phosphate, in the presence and absence of fructose 2,6 bisphosphate. Also illustrate the reaction velocity dependence on the concentration of ATP, with and without fructose 2,6 bisphosphate.
- 8 12. Describe the reaction catalyzed by glyceraldehyde-3-phosphate dehydrogenase in detail, including the structures of the three carbon intermediates, explaining how a molecule with high chemical energy is generated. Why does the product of this reaction have such a high free energy of hydrolysis?
- 8 13. Describe the biochemical steps in the generation and movement of an action potential down an axon. Why does the action potential only travel in one direction?
- 8 14. If the concentration of K^+ inside an erythrocyte is 140 mM and the concentration outside is 5 mM, what is ΔG for the transfer of 1 mol of K^+ from the outside to the inside? The temperature of the cell is $37^\circ C$ and the membrane potential is 0.07V, outside +ve. Would you expect to obtain a comparable value for the transport of Na^+ out of the cell? Why or why not?

END

Marks

- 20 1. Write brief notes on:
- Channeling of enzyme catalyzed reactions
 - ErbB
 - Proteoglycans
 - Edman degradation
 - Isozymes (provide an example)
- 10 2. Calculate the free energy required to transport 1 mol of hydrogen ions from the cytoplasm of parietal cells in the stomach lining of a live mammal into the lumen of the stomach. Assume that the pH of the parietal cell cytoplasm is 7 and that the pH of the gastric juice is 1. The parietal cell membrane potential is 0.1 V, outside positive. Show your calculations. Where could the energy to drive this transport come from?
- 15 3. a) Explain how and why changes in carbohydrate catabolism are different during the flight or flee response.
b) Carbohydrate metabolism does not change in erythrocytes during the flight or flee response. What reasons can you think of to explain this?
- 10 4. a) Many of the chemical groups known to organic chemists are not found in the building blocks of biomolecules. Why do you think this is the case?
b) Draw the structure of NADH, identifying the types of bonds that join its constituents together.
c) Why do you think that enzymes have evolved to use redox pairs like NAD⁺/NADH as electron carriers, rather than redox pairs such as acetaldehyde/ethanol?
- 8 5. Phosphofructokinase I (PFK I) is said to act as a valve, controlling the rate of glycolysis. Illustrate how and why this is so. It is not necessary to provide details of the modulators of PFK I.
- 8 6. a) When purifying an enzyme, why do you need to keep track of total activity? Specific

- activity?
- b) When you have finished a purification, how can you determine whether or not your protein is pure?
- 8 7. The enzyme phosphoglucomutase is a close relative of phosphoglyceromutase. Illustrate the likely reaction mechanism of phosphoglucomutase. Is ATP required by this enzyme?
- 15 8. Describe the acetylcholine receptor:
- a) As a transmembrane protein.
b) As an ion channel.
c) As a receptor in cell signaling.
- 8 9. a) Illustrate how phospholipases degrade glycerophospholipids. It is not necessary to provide the complete structure of the lipid you choose.
b) Which phospholipases may be involved in cell signalling, and what are the products of the reactions they catalyze?
- 8 10. You notice a slimy patch of goo on your carpet. It seems as if the goo is eating away at your carpet. Being an awesome biochemist, you figure out bacteria are feeding off your carpet. But wait, your carpet is made of nylon! How can this be? Perhaps the landfill and nuclear power plant next to your house have something to do with it...

You set out to determine how the bacteria can live off nylon since nothing known can. You scrape off some of the bug-infested carpet and take it to your lab. You culture large quantities of the bacteria and painstakingly purify a protein with the ability to cleave the nylon. You name the newly discovered enzyme Nylase. Nylon is a polymer made up of many repeating subunits (like the polysaccharides). It looks like this:

The squiggly lines at the ends indicate that this same unit is repeated many times in both directions. The arrow points to the bond that is cleaved to break up the nylon polymer.

- a) Just how good is the enzyme? If you just put nylon in water, the rate at which this bond will cleave is about 1 per year. In the presence of the enzyme, it's about 100 per second. What is the increase in the rate of the reaction?
- b) You make a solution that is 0.1 M in nylon. You add some enzyme and allow the reaction to reach equilibrium at 25 °C. You determine the concentration of nylon at equilibrium is 0.0001 M. What is the equilibrium constant for the reaction?
- c) What is the change in free energy ($\Delta G'^{\circ}$)?

END