

Instructions: This exam consists of 100 points on 6 pages. Please use the space provided to answer the question or the back of the preceding page. In questions with choices, all your answers will be graded and you will receive the best grade. Allot 1 min/2 points.

1. (3 pts) What fundamental principle allows one to determine the structure of proteins using X-ray diffraction techniques?

- The electrons surrounding the atom will scatter the x-rays (1 pt)
- The scattered X-rays will interfere with each other, and the interference depends on the position of the atoms. The interference can therefore be used to determine the position of the atoms (2 pts)

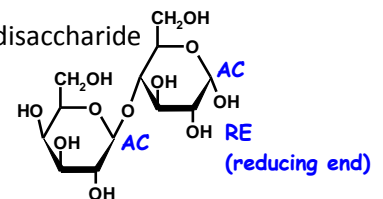
2. (2 pts) Fill in the blanks. The anomeric carbon is always involved in the formation of the glycosidic bond that joins two monosaccharides to form a disaccharide.

3. (4 pts) Fill in the blanks. Under conditions of low oxygen, yeast will convert pyruvate to ethanol in order to regenerate NAD⁺ for glycolysis.

4. (5 pts)

i) Indicate the reducing end with "RE" and one anomeric carbon with "AC" on the disaccharide shown on the right. (2 pts)

ii) Fill in the six blanks to complete the name of this sugar (3 pts)



β-galactopyranosyl -(1→ 4)- α glucopyranose

1 2 3 4 5 6

5. (6 pts) Please do one of the following two choices.

Choice A: Compare and contrast the structure and biological function of glycogen and cellulose. In what ways are they similar, and in what ways do they differ?

Choice B: Bacterial cell walls contain a polysaccharide component that is similar to cellulose.

- In what ways does the polysaccharide component in bacterial cell walls differ from cellulose?
- What additional component is found in bacterial cell walls?
- We possess an enzyme (lysozyme) that can digest bacterial cell walls, why is it not possible for us to digest cellulose?

Choice A:

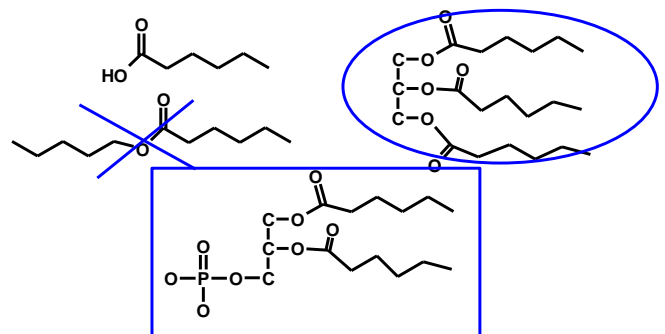
- Both are polymers containing glucose (2 pts)
- In glycogen the linkages are linear α(1-4) with α(1-6) branches. In cellulose they are β(1-4) linkages with no branching (3 pts)
- Glycogen is used for glucose storage, cellulose is a structural polysaccharide. (1 pt)

Choice B:

- The glucose units are replaced by alternating NAG and NAM residues - these are modified glucose (2 pts)
- The NAM units in the carbohydrate portion of each chain are linked together via a peptide (3 pts).
- Although lysozyme can hydrolyze β (1-4) linkages, it only binds to substrates that contain NAG - N-acetylglucosamine (1 pt)

6. (3 pts) Four compounds are shown on the right.

- Circle the triglyceride,
- Put a **box** around the phospholipid,
- Draw an 'X' through the wax.



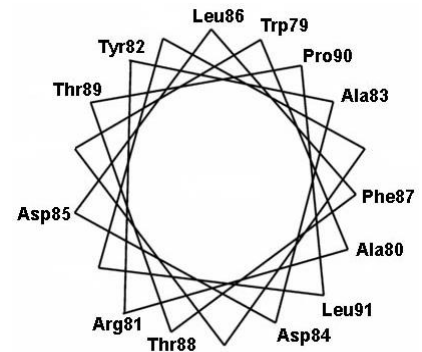
7. (6 pts) Please do **one** of the following four choices.

Choice A: What is the critical micelle concentration (CMC) and how would it depend on the number of carbons in a fatty acid?

Choice B: How does cholesterol affect the properties of membranes and why is this effect important for the function of biological membranes?

Choice C: In what way is the function of membrane transport proteins similar to soluble enzymes? In what ways do they differ?

Choice D: The diagram on the right is a helical wheel depiction of an α -helix in a membrane protein. Indicate which part of this helix is exposed to the lipids and what energetic features would stabilize this interaction? Briefly justify your answer (Hint: The plot on the last question may be helpful).



Choice A: It is the highest possible concentration of monomeric fatty acids in solution. Above this concentration any added fatty acids will form micelles. As the fatty acid length increases the CMC should decrease because the solubility will decrease.

Choice B: Cholesterol keeps the membrane fluid, permitting conformational changes to occur in membrane enzymes and the diffusion of electron carriers, such as CoQ.

Choice C: Both convert substrate to product. A soluble enzyme will typically cause a change in the chemical structure of the substrate. The transport protein doesn't change the structure, but binds the "substrate" on one side of the membrane and moves the compound across the membrane, producing the product. The only difference between substrate and product is the location.

Choice D: The right side is exposed to the lipid because it contains mostly non-polar residues. These will interact favorable with the lipids due to the hydrophobic effect and van der Waals.

8. (12 pts) Please do **one** of the following two choices.

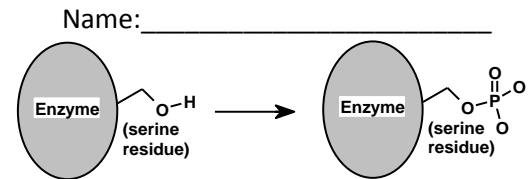
Choice A: Briefly explain why corn oil is a liquid at room temperature, but that corn oil margarine is a solid at room temperature. What fundamental thermodynamic interaction (e.g. H-bonds) is responsible for this difference?

Choice B: Explain why fatty acids form micelles in water while phospholipids forms bilayers. What fundamental thermodynamic interaction is responsible for the assembly of both of these structures?

Choice A: Corn oil contains *cis* unsaturated double bonds that cause a bend in the fatty acid chain, disrupting **van der Waals** interactions. Margarine is made by converting these double bonds to single bonds, producing saturated fats. The fatty acids pack together better, increasing van der Waals forces, raising the melting temperature.

Choice B: Fatty acids have a larger polar head group than the hydrocarbon chain, hence they are cone shaped and will pack together to form a spherical structure. The area of the polar head-group in a phospholipid is roughly equal to the two fatty acid chains, thus these pack together as cylinders. The assembly is driven by the **hydrophobic effect**.

9. (6 pts) Is the following reaction catalyzed by a kinase or a phosphatase? Briefly justify your answer and list any reactants or products that may be missing from the diagram.



- The reaction shown is catalyzed by a kinase, which transfers a phosphate group from ATP to the substrate. (5 pts)
 - The missing reactant is ATP, and the missing product is ADP. (1 pt)
10. (8 pts) Please do **one** of the following two choices. Be sure to discuss the sign of ΔG in your answer.
- Choice A:** The reaction fructose-6-P + $P_i \rightarrow$ fructose-1,6-P has an unfavorable standard energy ($\Delta G^\circ = +15$ kJ/mol). Why is this reaction spontaneous in glycolysis? What form of coupling is this?
- Choice B:** How can indirect coupling be used to insure a reaction in a pathway is spontaneous? Give an example from either glycolysis or the TCA cycle.

Choice A:

- ATP provides the phosphate group, i.e. it is transferred from ATP to F-6-P, generating F-1,6-P and ADP. The removal of a phosphate from ATP releases 30 kJ/mol, so the net ΔG° is -15 kJ/mol. (5 pts)
- This makes the Gibbs energy negative - so the reaction is spontaneous. (2 pts)
- This is direct coupling (1 pt)

Choice B: Indirect coupling involves maintaining the concentration of the product lower than its equilibrium concentration. This makes the term $RT\ln[B]/[A] < 0$, making the Gibbs energy less than zero, which makes the reaction spontaneous (7 pts)

- Glycolysis: The reaction catalyzed by aldolase has a large positive ΔG° which is offset by keeping the products very low because the last step in glycolysis is energetically favorable (1 pts)
- TCA cycle: The last oxidation step (fumarate to OAA) is unfavorable, however the next step in the reaction (citrate synthase) is very favorable and keeps the concentration of OAA very low (1 pts)

11. (11pts) Assume that you had a cream cheese bagel for breakfast.

i) Briefly discuss how the carbon atoms in the food are ultimately converted to CO_2 for any **two** of: a) fats, b) proteins, c) carbohydrates that were contained in the bagel. You only need to simply state the major pathways that would be involved in the production of CO_2 and the location of the pathway (6 pts).

a) Fats: Triglycerides \rightarrow fatty acids \rightarrow Activation (cytosol) \rightarrow oxidation (M. Matrix) \rightarrow TCA (M. Matrix).

b) Proteins \rightarrow amino acids \rightarrow TCA cycle (M. matrix).

c) Carbohydrates: Glucose \rightarrow glycolysis (cytosol) \rightarrow TCA (M. matrix).

ii) Why can't you convert most of the carbon in fats to sugars? (3 pts)

- Glucose is made from pyruvate in gluconeogenesis. Fatty acids are oxidized to acetylCoA and the carbons in acetyl CoA cannot be converted to pyruvate in humans because we lack the enzyme to do so.

iii) Complete **only one** (i.e. only one will be graded) of the following fill in the blanks: (2 pts)

- Prior to entering the TCA pathway, pyruvate is converted to AcetylCoA, using NAD⁺ as the electron acceptor. This reaction is catalyzed by a dehydrogenase (general name).
- In the TCA/fatty acid oxidation pathway, an alkane is converted to a alkene, using FAD as the electron acceptor. This reaction is catalyzed by a dehydrogenase (general name).
- In the TCA/fatty acid oxidation pathway, an alcohol is converted to a ketone, using NAD⁺ as the electron acceptor. This reaction is catalyzed by a dehydrogenase (general name).
- In the glycolysis pathway, an aldehyde is converted to a phosphate ester/carboxylic acid, using NAD⁺ as the electron acceptor. This reaction is catalyzed by a dehydrogenase (general name).

12. (12 pts) Please do **one** of the following two choices related to the conversion of captured energy to ATP. Feel free to use a diagram in your answer.

Choice A: Briefly describe the second to last step in the conversion of the energy in the bagel to ATP, i.e. electron transport. Be sure to indicate how energy released from this step is stored for the generation of ATP. Indicate the relative energy content of the two compounds that enter this pathway.

Choice B: Briefly describe the last step in the conversion of the energy in the bagel to ATP, i.e. the synthesis of ATP from the energy source generated in part A of this question.

Choice A:

The movement of electrons through the electron transport chain provides energy to move protons across the inner membrane, establishing a proton gradient – this is how the energy is stored (5 pts).

- NADH: The electrons from NADH follow this path: I → CoQ → III → Cytochrome C → IV → O₂. (3 pts)
- FADH₂: The electrons from FADH₂ follow this path: II → CoQ → III → Cytochrome C → IV → O₂. (3 pts)

Oxidation of NADH pumps more protons than FADH₂ (10 versus 6) (1 pt)

Choice B:

- The enzyme that synthesizes ATP (ATP synthase, ATPase) resides in the inner mitochondrial membrane and matrix.
- The part that is in the matrix contains a gamma subunit, 3 alpha, and 3 beta. (4 pts for general description of enzyme)
- The conformation of the beta subunits depends on their interaction with the gamma, and this interaction changes every time 3 H⁺ pass through the enzyme. This is due to rotation of the gamma subunit, 120 deg/3 H⁺. (4 pts for stating effect of gamma on conformation of beta subunits)
- The initial conformation has low affinity for ADP and ATP. Thus ATP formed in the previous cycle would be released.
- The second conformation has high affinity for ADP and Pi, so they bind.
- The third conformation is such that ATP is lower energy than ADP + Pi, so the bound ADP + Pi is spontaneously converted to ATP.
- Returning to the initial conformation will release the new ATP. (4 pts for description of mechanism).

13. (12 pts) Please do **one** of the following three choices. If appropriate you should state the type of regulation (feedback, product, allosteric) in your answer.

Choice A: Glycolysis, gluconeogenesis, and the TCA cycle are all regulated by “energy sensing”. Select **one** of the three pathways and describe:

- i) the step that is regulated (2 pts).
- ii) the compounds that regulate that step, and whether they activate or inhibit the step (4 pts).
- iii) why this form of regulation is useful to the cell (4 pts).

Choice B: The liver cell responds to a number of different hormones, including insulin, glucagon, and epinephrine. Select **any one** of these hormones and:

- i) State under what conditions the hormone would be released (e.g. low blood glucose levels) (1 pts).
- ii) Whether proteins become phosphorylated or not, and the general mechanism for this event (3 pt).
- iii) How the response to the hormone affects the synthesis and degradation of glycogen and why this response is appropriate (6 pts).

Choice C: Discuss coordinated regulation of glycolysis and gluconeogenesis, by hormonal regulation. Your answer should indicate why this form of regulation is useful to the cell.

Choice A:

- i) Glycolysis: PFK – Inhibited by ATP, activated by AMP & ADP (allosteric). Since glycolysis produces ATP, high levels should shut it off. AMP and ADP levels indicate that the cell needs ATP, so glycolysis should be on.
- ii) Gluconeogenesis: bis phosphatase – Inhibited by AMP. Since gluconeogenesis requires ATP (anabolic pathway), it should not operate unless there is sufficient ATP. Insufficient ATP would be indicated by high levels of AMP, turning the pathway off.

iii) TCA Cycle: Pyruvate dehydrogenase and citrate synthase are inhibited by ATP and NADH. High levels of these compounds indicate that the cell has enough energy, so the entry of carbon into the TCA cycle should be suppressed.

Choice B:

- Low blood glucose/glucose demand:
- Glucagon or epinephrine released.
- Enzymes become phosphorylated due to activation of G-proteins and adenylyl cyclase.
- Glycogen phosphorylase is active, releasing glucose from glycogen.
- This allows the liver to provide glucose to raise the blood glucose levels.
- High blood glucose
- Insulin released
- Binding of insulin to its receptors leads to dephosphorylation of enzymes
- Glycogen synthase is active when dephosphorylated, so glucose is incorporated into glycogen
- This allows the liver to store the excess glucose in glycogen.

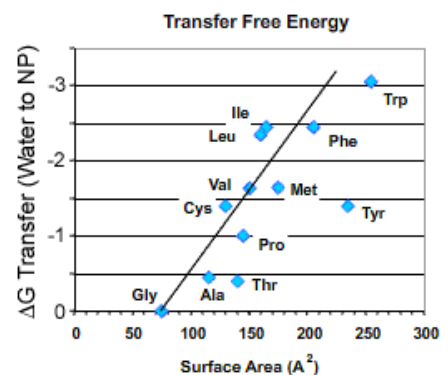
Choice C:

- F2,6 P levels are regulated by hormones.
- Low blood sugar - F2,6 P levels will be low, high blood sugar - F2,6 P levels will be high.
- High F26P levels activate PFK in glycolysis, this makes sense because there is plenty of glucose so glycolysis should be allowed to be on in case the liver cell needs to make ATP.
- High F26P levels inhibit the bisphosphatase in gluconeogenesis, so only one pathway is on.

14. (10 pts) Please do **one** of the following two choices.

Choice A: A hypothetical cell utilizes a chloride ion gradient to produce ATP. Assuming that the concentration of Cl^- outside the mitochondrial membrane is 100 mM, and the concentration is 10 mM inside, what is the minimum number of chlorides required to provide enough energy to synthesize one ATP? You can assume that the membrane potential is -100 mV, with the inside **negative**, and that $T=300\text{K}$.

Choice B: The concentration of two short polypeptides in membranes is measured. One polypeptide, consisting entirely of alanine residues is predominately found in the aqueous solution, while the other peptide, which consists of phenylalanine residues, is predominately found dissolved in the membrane. Explain, in **quantitative** terms, the different behavior of the two peptides. The diagram on the right may be helpful in your answer.

**Choice A:**

$$\Delta G = RT \ln \frac{[x]_{in}}{[x]_{out}} + ZF\Delta V = (8.3)(300) \ln \frac{10}{100} + (-1)(96,000)(-0.1)$$

$$\Delta G = -5.7 \frac{\text{kJ}}{\text{mol}} + 9.6 \frac{\text{kJ}}{\text{mol}}$$

The overall Gibbs energy is positive (~4 kJ/mol) therefore the ions will **not** flow spontaneously across the membrane, an additional 4 kJ/mol would be required to move them across the membrane. Thus there is no energy for the synthesis of ATP.

(5 pts for correct calculation, 5 pts for correct logic)

Choice B:

- The overall standard energy (ΔG°) for insertion of the peptide consists of a positive term for the mainchain atoms (hindering insertion) (+ 2 pts).
- This is offset by a negative contribution from the side chain. Alanine residues have a negative standard energy of -0.5. Since these don't insert, the contribution of the mainchain atoms has to be at least +0.5 (+4 pts)
- Phenylalanine sidechains have a larger favorable standard energy for transfer, this is sufficient to overcome the positive standard energy for the mainchain atoms. (+ 4 pts)

Bonus (2 pts): Why is it better to use maple syrup on your pancakes instead of syrup made from high fructose corn syrup? **Maple syrup contains mostly glucose, which enters glycolysis above the regulated step and thus its oxidation would be controlled. Fructose enters below the regulated step, so it would be oxidized to pyruvate (and potentially converted to fats) without significant metabolic control.**