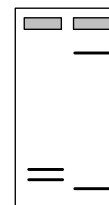


This Exam contains 14 questions and 100 pts in 6 pages. Budget 1 min/2 pts.

1. (8 pts) Please do **one** of the following two choices:

Choice A: A protein is treated with SDS and subject to polyacrylamide gel electrophoresis. In the stained gel on the right the unknown protein is in the **left** lane and the two standards, with molecule weights of 10 kDa and 100 kDa, are in the **right** lane. If the native molecular weight was determined to approximately 60 kDa, describe the quaternary structure of this protein.



Choice B: Briefly describe the process by which structures are determined using x-ray diffraction.

Choice A:

The protein contains two different subunits, both about 10 KDa in size since they migrate close to the 10 kDa standard (lower band), therefore the quaternary structure is $(\alpha\beta)_N$ where N is some integer. The molecular weight for N=1 will be ~20 kDa. The native molecule weight is 60 kDa, so N=3.

The quaternary structure is $(\alpha\beta)_3$.

Choice B:

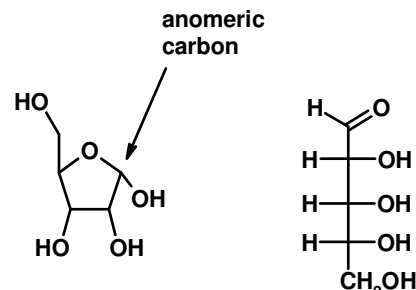
- i) proteins are crystallized.
- ii) X-rays are directed at the crystal.
- iii) Scattering of x-rays by the electrons occurs.
- iv) **Scattering depends on the relative atomic positions.**
- v) Intensity (and phase) of scattered X-rays are used to generate an electron density map.
- vi) Atoms are placed into regions of high electron density, giving a model of the structure.

2. (6 pts) A five carbon monosaccharide is shown on the right.

- a) is this compound an **aldose** or ketose?
- b) Draw the cyclic, or ring form of this sugar (you can omit hydrogens in your drawing.)

The -OH group on C4 will form a bond with C1

- c) Indicate the anomeric carbon on your drawing.
- d) Is this sugar glucose, **ribose**, or fructose?

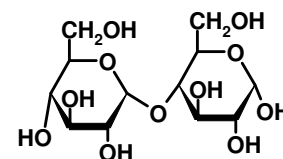


3. (2 pts) The disaccharide on the right is produced after partial hydrolysis of glycogen or starch. Correct the single error in its name:

β -glucopyranosyl-(1-4)- α -glucopyranose

Both anomers are pointing down, or in the α configuration. So the correct name is:

α -glucopyranosyl-(1-4)- α -glucopyranose



4. (8 pts) Please do **one** of the following two choices. You may answer the question with a sketch.

Choice A: Compare and contrast glycogen (or starch) to cellulose.

Choice B: Briefly describe the structure of bacterial cell walls.

Choice A:

Both are polymers of glucose (+3 pts)

Glycogen is composed of linear $\alpha(1-4)$ chains cross linked to other $\alpha(1-4)$ chains by $\alpha(1-6)$ linkages (+3 pts)

Cellulose consists of $\beta(1-4)$ linkages with no crosslinks. (2 pts)

Choice B:

Linear polysaccharide chains composed of modified glucose, NAM and NAG.

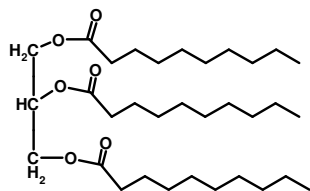
NAM and NAG alternate and are joined by $\beta(1-4)$ linkages.

Short peptide attached to each NAM residue.

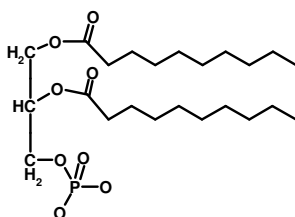
Peptides on adjacent strand crosslinked by another sort peptide.

5. (4 pts) The diagram on the right shows the partial structure of either a triglyceride or the phospholipid phosphatidic acid. Modify the diagram to complete the structure of **one** of the following two choices (please indicate your choice.)

Choice A: triglyceride.



Choice B: phosphatidic acid.



6. (10 pts) Please do **one** of the following two choices:

Choice A: The melting temperature of corn oil is approximately 10 C, while that of margarine which is made from the corn oil is about 30C. In what way was the corn oil modified to raise its melting temperature? Why did the melting temperature increase?

Choice B: The melting temperature of a series of phosphatidyl cholines is shown on the right, briefly explain this trend.

Choice A:

The cis double bond in the liquid corn oil are hydrogenated, by the addition of hydrogen (3 pts)

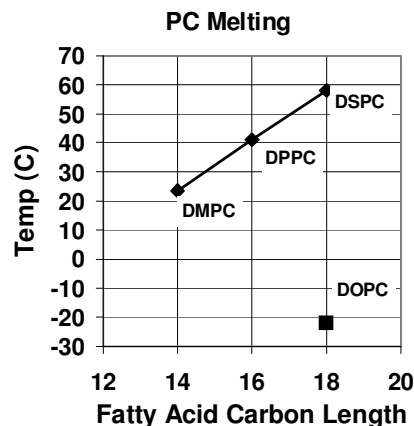
This produces either saturated bonds or tran-double bonds (2 pts)

This change causes the non-polar acyl chains to become straight, enhancing van der Waals contacts - leading to a higher melting temp (5 pts)

Choice B:

The top three points represent straight saturated acyl chains. The longer the chain, the higher the van der Waals interaction, the higher the Tm (5 pts)

The point at -20 C represents phospholipids with cis-double bonds. The cis double bond causes a kink in the acyl chain, disrupting van der Waals interactions (5 pts)



7. (12 pts) Please do **one** of the following two choices:

Choice A: Briefly describe the structural features of membrane proteins and energetic considerations that favor the insertion of the protein into the membrane. Your answer should discuss both mainchain as well as sidechain interactions.

Choice B: Fatty acids form micelles in solution while phospholipids form bilayers. .

- i) (8 pts) What is the major energetic force that causes these structures to form spontaneously? Briefly describe the molecular features of this force.
- ii) (2 pts) Briefly explain why different structures (micelles versus bilayers) are formed by the different compounds.
- iii) (2 pts) Explain why the CMC decreases as the length of the fatty acid increases.

Choice A:

The sidechains of membrane proteins that face the lipids are non-polar (4 pts).

The secondary structures are either helical or sheet, to insure that all H-bonds are formed in the folded protein because there are no donors or acceptors in the bilayer (6 pts)

The standard energy for burying the mainchain atoms is positive and unfavorable (1 pt)

The standard energy for burying non-polar sidechains is favorable. (1 pt)

Choice B:

i) The hydrophobic effect drives the assembly. The monomeric fatty acids or lipids will order water around their non-polar parts.

When micelles or bilayers are formed this water is released, increasing the entropy of the system. (8 pts)

ii) Fatty acids are cone shaped, with the wide part the acidic headgroup, so they pack in a spherical structure.

Phospholipids have an equal surface area for the polar head and non-polar tails, so they pack as cylinders, giving a planar bilayer. (2 pts)

iii) The CMC is the highest concentration of monomeric fatty acids. As the fatty acid becomes longer its solubility drops because of the hydrophobic effect. (2 pts)

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

Choice A: Select **either** carbohydrates **or** fatty acids and briefly discuss the metabolic fate of the carbons from these compounds. Your answer should state which pathways are used and approximately where the carbon is released as CO₂.

Carbohydrates enter glycolysis and then enter the TCA cycle. One CO₂ is released during oxidation of Pyruvate to acetyl-CoA and two more during the first part of the TCA cycle.

Fats are oxidized by fatty acid oxidation (β-oxidation) to generate acetyl-CoA units that enter the TCA cycle. The two carbons are released in the first part of the TCA cycle.

Choice B: A person on a high protein/fat diet cannot maintain high levels of strenuous exercise without running out of energy, however they are perfectly comfortable doing less strenuous activities. Explain the metabolic basis of this observation.

Strenuous exercise requires the rapid production of ATP from glucose using glycolysis.

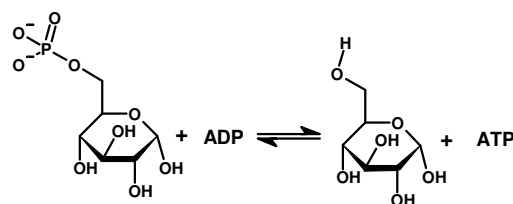
This glucose comes from glycogen.

An individual on a high protein/fat diet has low glycogen levels due to the fact that none of the carbons from fatty acids can be used to make glucose and only a small number of aminoacids can be converted to glucose.

9. (5 pts) Please do **one** of the following two choices.

Choice A: Is the reaction shown to the right catalyzed by a phosphatase, a kinase or a dehydrogenase? Briefly justify your answer.

Choice B: What is the major difference between a product inhibitor and a feedback inhibitor? Give an example of one and explain its importance in regulation.



Choice A:

This is a kinase because origin of the phosphate in the reverse reaction is ATP. The reverse reaction, $\text{Glucose} + \text{ATP} \rightarrow \text{G-6-P} + \text{ADP}$ is the first reaction in glycolysis, the enzyme is hexose kinase.

Choice B (4 pts for definition, 1 pt for example).

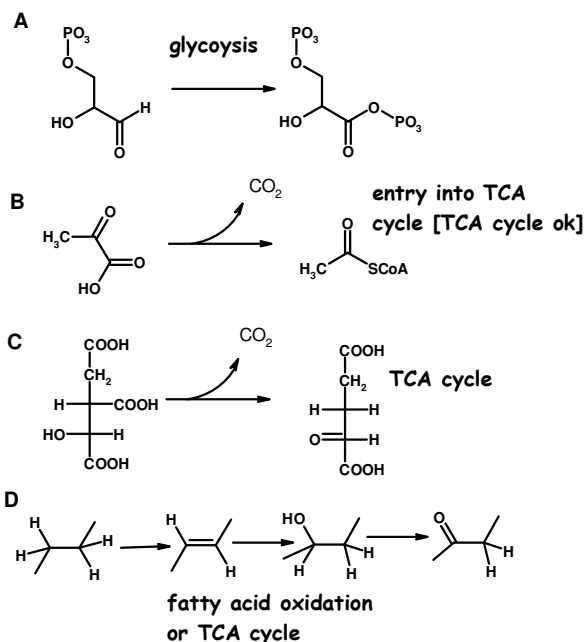
A product inhibitor is a product of the reaction and inhibits the enzyme by binding to the active site. Examples include the inhibition of pyruvate dehydrogenase by acetyl CoA or the inhibition of citrate synthase by citrate.

A feedback inhibitor is another compound from a down-stream step in the pathway. The inhibitor generally does not bind to the active site and is therefore an allosteric inhibitor. Examples include the inhibition of citrate synthase by succinyl-CoA or the inhibition of PFK by citrate.

10. (6 pts) A number of biochemical conversions are shown to the right:

i) (2 pts) Give the name of the pathway in which **one** of these reactions occurs. Write your answer next to the reaction.

ii) (4 pts) What is the common feature of all of these reactions? i.e. how have the left compounds been changed by the reactions? What compounds are not shown in these reactions?



These are all redox reactions, the right-hand compounds are more oxidized than the left [note that in the bottom series, on the first and last steps are redox reactions.] (3 pts)

The electron acceptors, NAD^+ and FAD , have been omitted. (1 pt)

11. (5 pts) The concentration of sodium ions (Na^+) outside a cell is 100 mM, while the concentration inside the cell is 10 mM. The voltage difference across the membrane is -100 mV. Will the transport of sodium occur spontaneously, or is the system at equilibrium?

$$\Delta G = RT \ln \frac{[\text{Na}^+]_{\text{IN}}}{[\text{Na}^+]_{\text{OUT}}} + ZF\Delta V$$

The first term is zero since the concentrations inside and out

are equal. The second term is negative since ΔV is negative, therefore sodium ions will spontaneously flow in since $\Delta G < 0$.

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

Choice A: The energy released by degradative pathways is directly captured on which types of compounds? Briefly explain how the energy on these compounds is converted to a hydrogen ion (proton) gradient across a membrane during electron transport.

Choice B: Briefly explain how the hydrogen ion gradient generated in choice A is converted to ATP by ATP synthase.

Choice C: Select **either** direct **or** indirect coupling and explain how coupling can be used to insure that a step in a metabolic pathway is spontaneous. How is the corresponding reaction in the reverse direction made to be spontaneous? Give one example to illustrate your answer.

Choice A: The energy is captured by the formation of reduced electron carriers, NADH and FADH₂. (2 pts)

The electron transport chain consists of (6 pts)

- four membrane bound complexes
- a non-polar electron carrier Q
- a water soluble carrier cytochrome C.

In the case of NADH the electrons go from complex I to Q to complex 3 to cytochrome C to complex 4. (2 pts for either a description of NADH or FADH₂ oxidation).

In the case of FADH₂ the electrons go from complex II to Q to complex 3 to cytochrome C to complex 4.

In both cases the electrons are used to reduce water.

Complex I, III, and IV all pump protons across the membrane. (2 pts).

Choice B: ATP synthase consists of two subunits. The one in the membrane (F_o) transports the protons, the one in the mitochondrial matrix (F₁) synthesizes ATP. (2 pts)

The F₁ domain contains three β-subunits, whose conformation depends on the relative orientation of the γ-subunit. The orientation of the γ-subunit changes by 120 degrees every time 3 protons are pass through the membrane. (6 pts)

This rotation causes the β-subunit to go through the following conformations: (4 pts)

- i. The first conformation has low affinity for ATP or ADP.
- ii. The second conformation binds ADP + Pi
- iii. The third conformation has an environment where ATP is more stable than ADP, so ATP forms.
- iv. Returning to the first conformation causes the release of the newly synthesized ATP.

Choice C:

Direct coupling. A reaction is made favorable by using the energy from a favorable reaction to drive an unfavorable one in the same active site. Example phosphorylation of glucose by hexose kinase or the phosphorylation of fructose-6-P by PFK.

In the reverse direction a different mechanism is used, e.g. simple hydrolysis of the phosphate group.

Indirect coupling. A reaction is made favorable by reducing the concentration of the products to below their equilibrium levels by having favorable steps further on in the pathway. This makes the $RT\ln[B]/[A]$ term negative. Example is aldolase.

Nothing special has to be done to reverse the pathway, just make the concentration of [A] below its equilibrium levels.

13. (4 pt) Briefly explain how glucose is converted to ethanol by yeast (3 pts). What environmental conditions would enhance the production of ethanol (1 pt)?

Pyruvate is converted first to acetylacetate, and then to ethanol, under conditions of low O₂ levels to regenerate NAD⁺ for glycolysis.

14. (10 pts) Please do only one of the following three choices.

Choice A: Briefly describe how energy sensing is used to regulate energy production in the cell. Your answer should provide one example of a step that is regulated by energy sensing.

Choice B: Select any **one** of the following three hormones: glucagon, or epinephrine, or insulin, and answer all of the following questions.

- Under what conditions (e.g. blood glucose levels) would your hormone be released?
- In the presence of your hormone, would the liver store glucose in glycogen or release glucose from glycogen?
- Briefly describe the molecular events that are responsible for the storage or release of glucose from glycogen under the influence of the hormone you have chosen. Your answer should include a brief discussion of changes in protein phosphorylation levels of the enzymes involved in glycogen synthesis and degradation.

Choice C: Select any **one** of the following three hormones: glucagon, or epinephrine, or insulin, and briefly describe how glycolysis and gluconeogenesis are regulated by this hormone. Your answer should indicate which steps in these pathways are regulated and the interplay between F2,6P levels, hormones, and the regulation of these steps.

Choice A:

Pathways that produce energy (glycolysis, TCA cycle) are turned off when the cell has high energy reserves, preventing the cell from oxidizing carbon when it doesn't need to (6 pts).

High levels of ATP inhibit PFK in glycolysis (4 pts)

High levels of ATP and NADH inhibit pyruvate dehydrogenase and citrate synthase.

Choice B:

glucagon	epinephrine	Insulin
Low blood sugar	request from the central nervous system to increase glucose levels in the blood.	High blood sugar.
Release glucose from glycogen		Store glucose in glycogen
- The binding of the hormone causes the activation of kinases via G-protein activation/adenyl cyclase activity - This leads to protein phosphorylation. - The enzyme that degrades glycogen (glycogen phosphorylase is active when phosphorylated)		- The binding of insulin activates protein phosphatases. - The enzyme that synthesizes glycogen from glucose is active with dephosphorylated.

Choice C:

Glucagon or epinephrine will shut down glycolysis to prevent the entry of glucose into glycolysis.

F26P levels fall due to the phosphorylation of the enzyme that degrades F26P (5 pts).

PFK is only active when F26P levels are high, so glycolysis is off.

bisPhosphatase, the enzyme in gluconeogenesis is inhibited by F26P, so the levels of F26P go low, it can be turned on to make additional glucose (5 pts).

Insulin will elevate the levels of F26P because the enzyme that makes F26P is active when dephosphorylated (5 pts).

F26P will activate PFK allowing glycolysis to proceed. Gluconeogenesis is turned off because the bisphosphatase is inhibited by F26P (5 pts).