7.014 Quiz I Handout

Quiz I announcements:

• Quiz I: Friday, March 10  12:05 - 12:55
  Walker Gym, 3rd floor  (room 50-340)

  **This will be a closed book exam**

• Quiz Review Session: Wednesday, March 8
  7:00 - 9:00 pm
  room 34-101

• Open Tutoring Session: Thursday, March 9
  4:00 - 6:00 pm
  room 66-144

• Below is a compilation of last year’s quiz problems that cover material on our Quiz I (solutions at the end). It should give you an idea of the format and types of questions we tend to ask on the exams. There is some difference in covered material and emphasis on various parts of the material between last year’s and this year’s versions of the course. We do not guarantee that this year’s quiz will cover the exact same subtopics.
Question 1 (36 points)

Below is a ribbon representation of the $K^+$ channel, a membrane spanning protein made up of four copies of a single polypeptide. The $K^+$ channel allows $K^+$ ions to be shuttled through the membrane.

a) What protein secondary structure is part of the $K^+$ channel protein as shown above?

b) Does the $K^+$ channel have quaternary structure? If yes, describe it.

c) Using $\text{膜}$. as a schematic of a phospholipid, draw a cross-section of the membrane around the $K^+$ channel shown above.

d) What type(s) of amino acids do you expect to find on the $K^+$ channel polypeptides

i) next to the tails of the membrane lipids? (Circle all that apply)

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<th>Polar</th>
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Why?

ii) next to the heads of the membrane lipids? (Circle all that apply)

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Why?
Question 1, continued

e) If you were trying to estimate the volume occupied by this protein, would the picture above provide all the information you need? Why or why not?

The positively charged K\(^+\) ion is a very small soluble molecule.

f) Explain why K\(^+\) cannot come across the membrane without a channel protein.

You isolate a mutant of the K\(^+\) channel that transports less K\(^+\) than normal. You run both the wild-type and mutant proteins on a denaturing gel and get the following result:

![Gel result]

\[\text{Wildtype} \quad \text{Mutant}\]

\[
\begin{array}{c}
\text{-} \\
\text{+}
\end{array}
\]

g) From the gel data we can conclude that (circle all that apply):

- shorter than
- longer than

i) Each subunit of the mutant protein is the same length as in the wild-type protein.

Justify your answer(s)

ii) The mutant and wild-type proteins could differ in their secondary structure.

Justify your answer(s)

primary

secondary

tertiary
Question 1, continued

The K\textsuperscript{+} channel has several binding pockets in which K\textsuperscript{+} ions may associate. Below is an image of one of the binding pockets of the K\textsuperscript{+} channel shown from above.

h) Circle the strongest type of interaction that exists between the K\textsuperscript{+} ion and the Glu residues.

Covalent bond  Van der Waals  Hydrogen bond  Ionic bond

i) You isolate a series of mutant K\textsuperscript{+} channel proteins where the two Asp residues have been replaced by amino acid X (see table below). For each X, indicate whether K\textsuperscript{+} binding in the resulting pocket will be stronger, weaker, or the same and give a brief explanation of your choice.

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j) Suppose you isolate another mutant that has four Glu residues instead of two Asp and two Glu residues in the pocket above. You find that the mutant has decreased K\textsuperscript{+} transport. Explain this result.
Question 2 (36 points)

a) Fill in the blanks:

A ________________ is an organism that is capable of making its own food store, while a ________________ must rely on getting food from the environment.

b) What types of organisms carry out

   i) glycolysis (circle all that apply)

      autotroph  heterotroph  prokaryotes  eukaryotes

   ii) photosynthesis (circle all that apply)

      autotroph  heterotroph  prokaryotes  eukaryotes

Below are simplified chemical flowcharts of glycolysis and the Krebs (citric acid) cycle.

In the course of glycolysis, NAD$^+$ is reduced to NADH.

c) In glycolysis, what is the original source of the electrons that are used to reduce NAD$^+$?
Question 2, continued

The glycolysis pathway produces energy from glucose.

d) In what molecule is this energy stored?

e) Where in that molecule is the energy that is used to perform work stored? Be specific.

Breakdown of the molecule (in d) is often coupled with other reactions in the cell, making the new, coupled, reaction proceed at an appreciable rate.

f) Describe one mechanism that is commonly used in such coupled reactions.

S. oxyphiliae is an organism that can undergo fermentation or respiration.

g) You take equal aliquots of the same oxyphiliae culture and supply both with equal amount of food. You place aliquot A into an airtight bottle and aliquot B into an open shallow container.
   i) Are the cells in cultures A and B deriving all of their energy in the same way? Explain.

   ii) Will one culture run out of food faster? If yes, state which culture, and explain why. If no, explain why not.

   iii) At the time when both cultures run out of food, will there be approximately the same number of cells in each? Why or why not?

h) In the Krebs cycle itself, only one ATP molecule is generated per molecule of pyruvate. Yet we know that respiration overall yields more additional ATP molecules. Beginning with the products of the Krebs cycle, briefly outline how that additional ATP is generated.
**Question 2, continued**

i) What is the main overall product of the dark reactions of photosynthesis?

All enzymatic reactions, including those in glycolysis and the Krebs cycle, are reversible. You decide to study where in the cell glycolytic enzymes are found. You are surprised to find that these enzymes are not only found in the cytoplasm, but also in chloroplasts.

j) Explain why these enzymes are found in chloroplasts.

k) Explain, in evolutionary terms, why two species that diverged billions of years ago still use many of the same enzymes and reactions.

---

**Question 3 (14 points)**

Below is the energy diagram for the reaction C+D → A+B.

![Energy Diagram](attachment:energy_diagram.png)

a) On the energy diagram above, label the following:

- $E_a (\Delta G^\ddagger)$
- $\Delta G$
- A+B
- C+D

b) Based on the diagram above,

- $\Delta G < 0$
- $\Delta G = 0$
- $\Delta G > 0$
**Question 3, continued**

c) How would the enzyme change

i) $\Delta G$?
Explain

ii) $E_a (\Delta G^\ddagger)$?
Explain

iii) speed of reaction?
Explain

**Question 4 (14 points)**

Barney is an alien. On his ship, hidden in the Stata building, you find alien bacteria that metabolize wood. You call this species *A. termiticus*, and call your original strain BLT (for “Barney’s little termiticus”).

You subject a sample of BLT to mutagens, and isolate a new strain that no longer metabolizes wood. You conclude that you have succeeded in disrupting at least one gene necessary to metabolize wood. You call the mutant strain M.

You mix a sample of M with a sample of heat-killed wild type BLT, and the resulting strain metabolizes wood. You summarize your data in the following table:

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</tr>
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<td>No</td>
</tr>
<tr>
<td>M</td>
<td>No</td>
</tr>
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<td>Heat-killed BLT +M</td>
<td>Yes</td>
</tr>
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</table>

a) Did the content of any of the BLT or M cells change in the experiment? If yes, which cells underwent the change, and what change occurred? If not, explain why there was no change.
Question 4, continued

You plan to characterize the alien genetic material. You start by breaking some *A. termiticus* cells open to determine their molecular composition. You find that they contain various small molecules, carbohydrates, lipids, and two other macromolecules, A and B.

In order to determine which macromolecule is the carrier of genetic information, you repeat your previous experiment, but include test tubes where you treat the sample of the heat-killed BLT with either an agent that destroys macromolecule A (A-ase) or macromolecule B (B-ase). You find the following results (including the repeat of your previous experiment in the first 4 lines):

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<td>Heat-killed BLT + M</td>
<td>Yes</td>
</tr>
<tr>
<td>A-ase treated heat-killed BLT + M</td>
<td>Yes</td>
</tr>
<tr>
<td>B-ase treated heat-killed BLT + M</td>
<td>No</td>
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b) Which molecule is the carrier of genetic information in *A. termiticus*? Justify your answer.

Next, you set out to determine the structure of the alien genetic material molecule. You first determine that it has six types of bases that you name S, V, W, X, Y, Z. You further determine that the alien cell’s content of S is the same as its content of each of X and Z; and that the content of V is the same as its content of each of W and Y.

When you determine the structure of this molecule by X-ray crystallography, you are not surprised to find that the molecule consists of 3 interacting strands.

c) What base interaction combinations do you expect for this molecule?
STRUCTURES OF AMINO ACIDS
at pH 7.0

ALANINE (ala)
ARGININE (arg)
ASPARAGINE (asN)
ASPARTIC ACID (asp)
CYSTEINE (cys)
GLUTAMIC ACID (glu)
GLUTAMINE (glN)
GLYCINE (gly)
HISTIDINE (his)
ISOLEUCINE (ile)
LEUCINE (leu)
LYSINE (lys)
METHIONINE (met)
PHENYLALANINE (phe)
PROLINE (pro)
SERINE (ser)
THREONINE (thr)
TRYPTOPHAN (trp)
TYROSINE (tyr)
VALINE (val)
Solutions
Question 1 (36 points)

Below is a ribbon representation of the K^+ channel, a membrane spanning protein made up of four copies of a single polypeptide. The K^+ channel allows K^+ ions to be shuttled through the membrane.

k) What protein secondary structure is part of the K^+ channel protein as shown above? 

*K^+ channel contains only α-helices.*

l) Does the K^+ channel have quaternary structure? If yes, describe it.

*Yes, the channel has quaternary structure. It consists of four interacting polypeptides. It appears that each polypeptide interacts with its two neighboring polypeptides, such that the overall shape of the channel is a barrel.*

m) Using as a schematic of a phospholipid, draw a cross-section of the membrane around the K^+ channel shown above.

n) What type(s) of amino acids do you expect to find on the K^+ channel polypeptides
j) next to the tails of the membrane lipids? (Circle all that apply)

- [ ] Polar
- [ ] Nonpolar
- [ ] Positively charged
- [ ] Negatively charged

Why?
The tails of the membrane lipids are hydrophobic, and it is energetically favorable for the protein to have hydrophobic amino acids in the hydrophobic environment.

ii) next to the heads of the membrane lipids? (Circle all that apply)

- [ ] Polar
- [ ] Nonpolar
- [ ] Positively charged
- [ ] Negatively charged

Why?
The heads of the membrane lipids have negatively charged phosphate groups, and it is energetically favorable for the protein to have positively charged amino acids that can form ionic bonds, or polar amino acids that can form hydrogen bonds in that environment.
Question 1, continued

o) If you were trying to estimate the volume occupied by this protein, would the picture above provide all the information you need? Why or why not?
   No, the picture above is of a ribbon representation, i.e. backbone trace. It does not show any of the sidechains, and thus does not give us a clear picture of how much space the protein occupies. In addition, for the atoms in the backbone, the vdW radii are not shown.

The positively charged K\(^+\) ion is a very small soluble molecule.

p) Explain why K\(^+\) cannot come across the membrane without a channel protein.
   The tails of the membrane lipids are hydrophobic. It is not energetically favorable for a charged molecule, such as K\(^+\) to be in a hydrophobic environment. Thus, K\(^+\) by itself can not come across the membrane.

You isolate a mutant of the K\(^+\) channel that transports less K\(^+\) than normal. You run both the wild-type and mutant proteins on a denaturing gel and get the following result:

\[
\begin{array}{c|c}
\text{Wildtype} & \text{Mutant} \\
\hline
- & \text{Shorter than} \\
+ & \text{Longer than}
\end{array}
\]

q) From the gel data we can conclude that (circle all that apply):

j) Each subunit of the mutant protein is \underline{the same length as} in the wild-type protein.

Justify your answer(s)

The mutant protein shows only one band on the gel, meaning that it consists of only one type of subunit. Denaturing gel separates protein fragments based on their size, with shorter fragments traveling further. Since the mutant subunit travels farther on the gel, we conclude that the mutant subunits are shorter than the wild type ones.

ii) The mutant and wild-type proteins could differ in their \underline{secondary} structure.

Justify your answer(s)

Because the mutant subunits are shorter than wild-type subunits, it is clear that the primary structure is affected. If the lost fragment is of significant length, tertiary structure will most
Question 1, continued

The K⁺ channel has several binding pockets in which K⁺ ions may associate. Below is an image of one of the binding pockets of the K⁺ channel shown from above.

![Binding Pocket Image]

r) Circle the strongest type of interaction that exists between the K⁺ ion and the Glu residues.

Covalent bond  Van der Waals  Hydrogen bond  Ionic bond

s) You isolate a series of mutant K⁺ channel proteins where the two Asp residues have been replaced by amino acid X (see table below). For each X, indicate whether K⁺ binding in the resulting pocket will be stronger, weaker, or the same and give a brief explanation of your choice.

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<td>stronger</td>
<td>Same size, weaker interaction — H-bond instead of ionic bond</td>
</tr>
<tr>
<td>Leu</td>
<td>stronger</td>
<td>Approximately same size, weaker interaction — vdW instead of ionic bond</td>
</tr>
<tr>
<td>Phe</td>
<td>stronger</td>
<td>Weaker interaction — vdW instead of ionic bond. Phe is also significantly larger than Asp, so steric hindrance is likely</td>
</tr>
</tbody>
</table>

**t)** Suppose you isolate another mutant that has four Glu residues instead of two Asp and two Glu residues in the pocket above. You find that the mutant has decreased K⁺ transport. Explain this result.

*Glu is a negatively charged amino acid, just like Asp. Therefore, mode of interaction, i.e. ionic bond would be preserved. The only difference between Glu and Asp is the extra CH₂ group in Glu. Since the function of the protein is impaired, we conclude that it is harder for the K⁺ ion to fit into the new, smaller binding pocket.*
Question 2 (36 points)

a) Fill in the blanks:

An autotroph_________ is an organism that is capable of making its own food store, while a heterotroph_________ must rely on getting food from the environment.

b) What types of organisms carry out

   i) glycolysis (circle all that apply)

      autotroph   heterotroph   prokaryotes   eukaryotes

   ii) photosynthesis (circle all that apply)

      autotroph   heterotroph   prokaryotes   eukaryotes

Below are simplified chemical flowcharts of glycolysis and the Krebs (citric acid) cycle.

In the course of glycolysis, NAD$^+$ is reduced to NADH.

c) In glycolysis, what is the original source of the electrons that are used to reduce NAD$^+$?

The original source of electrons in glycolysis is glucose.
**Question 2, continued**

The glycolysis pathway produces energy from glucose.

d) In what molecule is this energy stored?  
ATP

e) Where in that molecule is the energy that is used to perform work stored? Be specific.  
In ATP, the energy used to do work is stored in the phosphate bonds.

Breakdown of the molecule (in d) is often coupled with other reactions in the cell, making the new, coupled, reaction proceed at an appreciable rate.

f) Describe one mechanism that is commonly used in such coupled reactions.

One common mechanism involves transferring the third phosphate of the ATP onto one of the reactants, thus raising its energy state such that the new, coupled reaction is now favorable. Another mechanism involves performing ATP hydrolysis on the same enzyme, but in a different active site than the reaction being catalyzed. In this mechanism, ATP hydrolysis often causes the enzyme to change conformation, allowing it to catalyze the main reaction.

*S. oxyphiliae* is an organism that can undergo fermentation or respiration.

k) You take equal aliquots of the same *oxyphiliae* culture and supply both with equal amount of food. You place aliquot A into an airtight bottle and aliquot B into an open shallow container.

i) Are the cells in cultures A and B deriving all of their energy in the same way? Explain.

No. Culture A derives all of its energy from glycolysis, while culture B derives additional energy (ATP) from Krebs cycle and oxidative phosphorylation.

ii) Will one culture run out of food faster? If yes, state which culture, and explain why. If no, explain why not.

Yes. Because culture A derives all of its energy from glycolysis, it only produces 2 molecules of ATP/molecule of glucose. Thus, it will have to utilize more glucose to perform same amount of cellular functions as culture B that derives 36 molecules of ATP/molecule of glucose through respiration.

iii) At the time when both cultures run out of food, will there be approximately the same number of cells in each? Why or why not?

No. Because culture B utilizes its food stores more efficiently, it will be able to maintain cellular functions for longer on the same amount of food. Therefore, it will have more time to divide, and it will have more cells than culture A.

l) In the Krebs cycle itself, only one ATP molecule is generated per molecule of pyruvate. Yet we know that respiration overall yields more additional ATP molecules. Beginning with the products of the Krebs cycle, briefly outline how that additional ATP is generated.

Krebs cycle is run in mitochondria, where e⁻ rich NADH and FADH₂ unload electrons onto the electron transport chain (ETC). As electrons are passed down the ETC, protons are pumped into the space between the inner and outer membrane, forming gradient. Because of the gradient, it is favorable for H⁺ to get back across the membrane. That proton motive force powers ATP synthase to add a P, to an ADP, creating ATP.
Question 2, continued

m) What is the main overall product of the dark reactions of photosynthesis?

The overall reaction of photosynthesis is \(6\text{CO}_2 + 6\text{H}_2\text{O} \rightarrow \text{C}_6\text{H}_{12}\text{O}_6\text{(glucose)} + 6\text{O}_2\), and the main product is glucose.

All enzymatic reactions, including those in glycolysis and the Krebs cycle, are reversible. You decide to study where in the cell glycolytic enzymes are found. You are surprised to find that these enzymes are not only found in the cytoplasm, but also in chloroplasts.

n) Explain why these enzymes are found in chloroplasts.

In the process of generating glucose from \(\text{CO}_2\), the cell runs some of the reactions of the glycolytic pathway in reverse. Because all enzymatic reactions are reversible, enzymes used to run the pathway backward in chloroplasts are the same that are used to run glycolysis in cytoplasm.

o) Explain, in evolutionary terms, why two species that diverged billions of years ago still use many of the same enzymes and reactions.

Once a pathway is developed, any mutant \((M)\) of the pathway that accidentally arises is likely to be less efficient. Even if some descendant of \(M\), \(M1\), if it was to acquire another mutation, might end up being significantly better than the original non-mutant organism, \(M\) itself is not likely to survive or generate enough progeny to allow \(M1\) to ever appear. Thus, evolutionary pressure works against changes to key pathways, ensuring significant conservation throughout evolution.

Question 3 (14 points)

Below is the energy diagram for the reaction \(\text{C} + \text{D} \rightarrow \text{A} + \text{B}\).

![Energy Diagram](image)

\[ \Delta G \approx 0 \]

\[ \Delta G < 0 \]

\[ \Delta G > 0 \]

d) On the energy diagram above, label the following:

- \(E_a(\Delta G^\ddagger)\)
- \(\Delta G\)
- \(\text{A} + \text{B}\)
- \(\text{C} + \text{D}\)

e) Based on the diagram above,

\[ \Delta G < 0 \]
\[ \Delta G = 0 \]
\[ \Delta G > 0 \]
Question 3, continued

f) How would the enzyme change

i) ΔG?

Explain

ΔG is unaffected by the presence of an enzyme. This is because it is a thermodynamic property of the reaction, and is determined by the amount of energy available to perform the work of converting reactants to products and vice versa. Since the energy is stored in the bonds, it is independent of the path the reaction takes, or the rate at which it occurs.

ii) E_a (ΔG‡)?

Explain

Activation energy will be lower in the presence of the enzyme. Enzymes lower the activation energy of the reaction, allowing it to proceed.

iii) speed of reaction?

Explain

The rate of reaction is affected. Reactants reach the transition state due to random fluctuations in energy caused by molecular motion. If E_a is lowered, much less energy is required to reach it, so more molecules will be able to do so, and the rate of the reaction will increase.

Question 4 (14 points)

Barney is an alien. On his ship, hidden in the Stata building, you find alien bacteria that metabolize wood. You call this species *A. termiticus*, and call your original strain BLT (for “Barney’s little termiticus”).

You subject a sample of BLT to mutagens, and isolate a new strain that no longer metabolizes wood. You conclude that you have succeeded in disrupting at least one gene necessary to metabolize wood. You call the mutant strain M.

You mix a sample of M with a sample of heat-killed wild type BLT, and the resulting strain metabolizes wood. You summarize your data in the following table:

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d) Did the content of any of the BLT or M cells change in the experiment? If yes, which cells underwent the change, and what change occurred? If not, explain why there was no change.

Some cells of type M underwent the change. They acquired some of the genetic material from the heat-killed BLT cells. In particular, some part of the genetic material they acquired encoded for the gene that was mutated in M. Having received this gene, M cells now behave like the wild-type BLT cells because they can now produce the agent (like the Earth proteins) that restores the wood metabolism pathway.
Question 4, continued

You plan to characterize the alien genetic material. You start by breaking some *A. termiticus* cells open to determine their molecular composition. You find that they contain various small molecules, carbohydrates, lipids, and two other macromolecules, A and B.

In order to determine which macromolecule is the carrier of genetic information, you repeat your previous experiment, but include test tubes where you treat the sample of the heat-killed BLT with either an agent that destroys macromolecule A (A-ase) or macromolecule B (B-ase). You find the following results (including the repeat of your previous experiment in the first 4 lines):

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</tr>
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<td>B-ase treated heat-killed BLT + M</td>
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e) Which molecule is the carrier of genetic information in *A. termiticus*? Justify your answer.

*B* is the carrier of genetic information in the alien organism. This is because when the BLT cells are treated with B-ase, no B remains in the sample, and, as a result, no transformation occurs. On the other hand, treatment with A-ase has no bearing on the transformation ability of the heat-killed BLT cells. We, therefore, must conclude that B is the carrier of genetic information.

Next, you set out to determine the structure of the alien genetic material molecule. You first determine that it has six types of bases that you name S, V, W, X, Y, Z. You further determine that the alien cell’s content of S is the same as its content of each of X and Z; and that the content of V is the same as its content of each of W and Y.

When you determine the structure of this molecule by X-ray crystallography, you are not surprised to find that the molecule consists of 3 interacting strands.

f) What base interaction combinations do you expect for this molecule?

*Based on the data, we expect S on one strand to interact with X on another strand and Z on the third strand. We would also expect V on one strand to interact with W on another strand and Y on the third strand.*