Next Step Public Office Hours: Biochemistry Content and Difficult Passage Review

• Welcome to Office Hours!
• Introduction
• What Do I Need for this Session?
• Biochem Content Review
• Difficult Passage
• What Next?
Introduction to Office Hours

• Thanks for coming to Next Step Office Hours!
• If you haven’t been here before, here’s how it works...
• These sessions are meant to be:
  Interactive
  Problem-focused
  Specific to your needs (so ask questions!)
• Today’s focus: review of biochemistry
• Future sessions: content review, FL review
• This is NOT a lecture! You can benefit most by:
  Raising your hand and speaking
  Commenting in the question box
  Responding to poll questions

Before Getting Started
1. If you have a microphone, make sure it is turned on and easily available.
2. Locate the hand-raise button on the toolbar on your screen.
3. Locate the Q box on the toolbar.
4. Let me know if you’re having any technical issues!

Think of your question after Office Hours are over?

• Post on the forums!
  forum.nextstepmcat.com
Biochem Content Review

Today, let’s focus on how to make the most of your biochemistry prep:

• **Overall study strategies**
  
  *Active learning*
  *Big-picture perspective*
  *Test-like thinking*

• **High-yield topics**
  
  *Enzymes & enzyme kinetics*
  *Glycolysis*
  *Krebs cycle*
  *Electron transport chain*
Biochem Study Strategies

Recurring theme for biochem:

*Don’t miss the forest for the trees!*

When studying, ask yourself ...

- Why does this matter physiologically?
  - Biomolecules: how does chemical structure connect to biological function?
  - Pathways: what does a pathway do?
- What are the inputs & outputs of a pathway?
- How is a pathway regulated (big-picture?)
- Does a pathway have any especially important steps?
Biochem Study Strategies

A big-picture approach to biochem:

How is biochem tested on the MCAT? How do you get the most bang for your buck in terms of studying?

Focus on:

• Principles

• Physiological function

• Interconnections with other subject matter
  
  Amino acids & acid-base chemistry
  Carbohydrates & stereochemistry
  Metabolism & physiology

What have your biochem experiences been like? What strategies work for you?
Enzymes and Enzyme Kinetics

What do enzymes do?

- **Enzymes are biological catalysts.**
- **Enzymes reduce activation energy of rxn.**
- **Reduced activation energy \( \rightarrow \) faster rate**
- **What do enzymes NOT do?**
- **Major types of enzymes:**
  
  | Oxidoreductases | Lyases |
  | Transferases   | Isomerases |
  | Hydrolases     | Ligases   |
Enzymes and Enzyme Kinetics

Michaelis-Menten saturation curve

\[ V_{\text{max}}: \text{the maximum rate of the reaction} \]

\[ K_m: \text{the amount of substrate needed for the enzyme to work half as fast as it can} \]

Why is \( K_m \) a useful thing to measure?

What are units for \( K_m \)? What about \( K_{\text{eq}} \) and rate constant \( k \)?

What other assumptions do MM curves make? (Hint: [enzyme])

Office Hours: Biochem Content and Difficult Passage Review
Enzymes and Enzyme Kinetics

Lineweaver-Burk plot

Why use LB plots?

- $V_{max}$ and $K_m$ can be more precisely determined.
- Types of inhibition can be visualized more clearly.

Remember, info about [substrate] is still on x-axis, and info about rate is still on y-axis.
Types of inhibition: understand first, memorize $K_m$ and $V_{max}$ effects second!

**Competitive inhibition:** inhibitor binds at active site

$V_{max}$ unchanged & $K_m$ increased: why?

**Noncompetitive inhibition:** inhibitor binds at allosteric site

$V_{max}$ reduced & $K_m$ unchanged: why?

**Uncompetitive inhibition:** inhibitor binds E-S complex

$V_{max}$ reduced & $K_m$ reduced: why?
Enzymes and Enzyme Kinetics

**Competitive**

- $V_{max}$ (uninhibited)
- $V_{max}$ (inhibited)
- $K_m$ (uninhibited)
- $K_m$ (inhibited)

**Noncompetitive**

- $V_{max}$ uninhibited
- $V_{max}$ inhibited
- $K_m$ inhibited
- $K_m$ Uninhibited

**Uncompetitive**

- Noncompetitive inhibition
- Competitive inhibition
- Uninhibited
- Uncompetitive inhibition

Office Hours: Biochem Content and Difficult Passage Review
1. A biochemist is investigating a reaction featured in human metabolism. He notes that all necessary reactants are present in his test tube and that the process should proceed spontaneously given the conditions, but observes that no products are being made. The most likely explanation is:

A) the rate of the uncatalyzed reaction is extremely slow.
B) a particular enzyme within live cells changes the mechanism of the reaction.
C) catalysts within human cells increase the amount of free energy released during the course of the reaction.
D) A and B only.

2. Catalase is an enzyme found in especially high concentrations in the liver. This molecule catalyzes the conversion of the reactive oxidative species hydrogen peroxide into water and oxygen. In the presence of this enzyme:

A) the conversion of hydrogen peroxide to water and oxygen gas is made spontaneous.
B) the rate of conversion of hydrogen peroxide to water and oxygen gas is increased.
C) the rate of conversion of water and oxygen gas to hydrogen peroxide is increased.
D) more than one of the above.
3. Priya is investigating the function of Enzyme D, which has a $K_m$ value of 0.175 mM. She adds a large quantity of competitive inhibitor (Compound G) into her test tube. Which of the following is most likely to be the apparent $K_m$ value that she subsequently observes?

A) 0.09 mM  
B) 0.10 mM  
C) 0.175 mM  
D) 0.500 mM

4. Which of the following changes may impact the $V_{max}$ or maximal reaction rate?

A) Altering the amount of enzyme  
B) Altering the amount of a noncompetitive inhibitor  
C) Altering the amount of a mixed inhibitor  
D) All of the above
Glycolysis

• Breakdown of glucose into pyruvate
  • Net ATP production: 2 molecules
  • 2 NADH molecules are also produced

• Anaerobic

• Products fed into....
  • Citric acid cycle
  • Fermentation
• Occurs in cytosol

• Tightly regulated to avoid futile cycle with gluconeogenesis

\[
\text{C}_6\text{H}_{12}\text{O}_6 + 2 \text{NAD}^+ + 2 \text{ADP} + 2 \text{P}_i \rightarrow 2 \text{pyruvate} + 2 \text{NADH} + 2 \text{H}^+ + 2 \text{ATP} + 2\text{H}_2\text{O}
\]
Glycolysis

- **Input**: Steps 1-5
- **Output**: Steps 6-10
- **ATP input**: Steps 1, 3
- **ATP output**: Steps 7, 10
- **Regulatory points**:
  - Step 1 (hexokinase)
  - Step 3 (phosphofructokinase [PFK])
  - Step 10 (pyruvate kinase)
- **Committed step**: Step 3
  - **PFK1 inhibited** by ↑ ATP, ↓ pH, and ↑ PEP (a downstream product)
  - **PFK1 activated** by ↑ AMP and fructose 2,6-bisphosphate
- **Rate of glycolysis**:
  - ↑ when [ATP] is low
  - ↓ when [ATP] is high
Gluconeogenesis

• Creation of glucose from pyruvate, lactate, glycerol (lipids), some AAs

• Occurs mainly in liver (+ cortex of kidneys).

• Different from glycolysis in 3 major steps
  • If not “bypassed” would be highly endergonic.

• Fasting conditions promote gluconeogenesis
  • Acetyl-CoA (from beta-oxidation): ↑ gluconeogenesis
  • Glucagon → inhibits glycolytic enzymes, promotes PEPCK → ↑ gluconeogenesis
  • High [AMP] promotes gluconeogenesis.
  • High [ATP] inhibits gluconeogenesis.
5. All of the following are true about glycolysis EXCEPT that:

A) it occurs in the cytoplasm.
B) it is an anaerobic process.
C) it results in a net production of 4 ATP.
D) it requires ATP in order to be initiated.

6. An aerobic organism is placed in an oxygen-poor environment and begins to produce:

A) an excess of acetyl-CoA.
B) an excess of NADH.
C) an excess of ATP.
D) an excess of ADP.
7. Of the following, which accurately describes a difference between glycolysis and gluconeogenesis?

A) Glycolysis can occur in all cells, while gluconeogenesis cannot.
B) Glycolysis and gluconeogenesis occur in different parts of the cell.
C) Only gluconeogenesis requires ATP to function.
D) Glycolysis can occur in the absence of oxygen, while gluconeogenesis cannot.

8. The end goal of gluconeogenesis is the:

A) formation of acetyl-CoA for use in the citric acid cycle.
B) creation of ATP to facilitate the continuation of biological processes.
C) regeneration of NAD+ to allow glycolysis to continue.
D) regulation of blood glucose levels.
Krebs Cycle

• Indirectly produces many ATP.
  • 3 NADH, 1 FADH₂, 1 GTP

• Cannot proceed anaerobically, BUT does not directly require O₂: why?

• Occurs in mitochondria.

• Pyruvate → acetyl-CoA occurs via the PDC
  • Acetyl-CoA ↔ pyruvate, fatty acids, ketogenic AAs, etc.

• Irreversible steps:
  1. Acetyl-CoA + oxaloacetate + H₂O → citrate + CoA-SH
     2 C 4 C 6 C
  3. Isocitrate + NAD⁺ → α-ketoglutarate + NADH + CO₂ (rate-limiting step!)
     6 C 5 C
  4. α-ketoglutarate + NAD⁺ + CoA-SH → succinyl-coA + NADH + CO₂
     5 C 4 C
Krebs Cycle

- Final step: L-malate + NAD$^+$ → oxaloacetate + NADH + H$^+$
  - Not thermodynamically favorable: why does it proceed?
  - Note application of Le Châtelier’s Principle & connection to Gen Chem!

- Regulated at 4 major points:  
  
  **Basic principle: negative feedback!**

  - At the PDC (upstream of cycle itself)
    - ↑ ATP, NADH, acetyl-CoA → PDK inactivates pyruvate dehydrogenase → ↓ Krebs cycle
  - Step 1: inhibited by citrate [direct product], ATP, NADH
  - Step 3: inhibited by succinyl-CoA and NADH [products], ATP
  - Step 4: inhibited by succinyl-CoA, NADH, ATP
9. During the Krebs cycle, isocitrate and NAD\(^+\) come together to form alpha-ketoglutarate, NADH and H\(^+\). Which of the following describe this type of chemical reaction?

A) Dehydration  
B) Hydration  
C) Decarboxylation  
D) Oxidation

10. In the Krebs cycle, NAD\(^+\) and FAD react with substrates to form NADH and FADH\(_2^+\). Over the same period of time that nine molecules of FADH\(_2^+\) are produced, how many units of NADH will be synthesized?

A) 9 molecules  
B) 18 molecules  
C) 27 molecules  
D) 36 molecules
11. High levels of acetyl-CoA stimulate which of the following enzymes?

I. Citrate synthase
II. Pyruvate carboxylase
III. Pyruvate dehydrogenase

A) I only
B) I and II
C) II and III
D) I, II, and III

12. Passage of one molecule of pyruvate through the Krebs cycle yields how many molecules of ATP when followed through oxidative phosphorylation?

A) 13
B) 15
C) 30
D) 36
Electron Transport Chain

General information:
- Occurs along inner mitochondrial membrane
- Provides energy for ATP synthase (proton gradient)
- ~30 ATP per glucose

Mechanism:
- 4 membrane-bound complexes transfer e⁻ from NADH and FADH₂ and pump H⁺ into intermembrane space
- \([H^+]_{\text{intermembrane space}} > [H^+]_{\text{matrix}}\)
- Protons rush down gradient, powering ATP synthase

Regulation:
- Inhibited by ↓ O₂, ↓ [ADP]/[ATP], ↓ NADH
- Stimulated by ↑ O₂, ↑ [ADP]/[ATP], ↑ NADH

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Image adapted from Fvasconcellos [Public Domain]
13. Which of these statements regarding proton pumping is accurate?

A) The intermembrane space becomes positive due to the protons being pumped into it.
B) The mitochondrial matrix becomes positive due to the protons being pumped out of it.
C) The intermembrane space becomes negative due to the protons being pumped out of it.
D) The mitochondrial matrix becomes negative due to the protons being pumped into it.

14. All of the following statements describe the net result of oxidative phosphorylation EXCEPT:

A) NADH and FADH$_2$ are oxidized to generate a proton gradient.
B) Controlled proton flow through ATP synthase is used to regenerate ATP.
C) Oxygen is the final electron acceptor in the electron transport chain.
D) The products of oxidative phosphorylation are retained for the next round of cyclic phosphorylation.
Amino Acids

**MCAT musts for amino acids**

- Understand chemical structure (peptide bond, etc.)
- Which amino acids have which properties?
- 3-letter abbreviations
- Specific structures and 1-letter abbreviations
- $pK_a$, acid-base properties
- How are amino acid properties & protein properties connected? Which residues would you expect in the transmembrane domain of a protein?

**Special amino acids (why?)**

- Cysteine
- Proline
- Glycine
Amino Acids

Acidic vs. basic functional groups

- **Acidic:** -COOH, -OH (Tyr), -SH (Cys)
- **Basic:** amines, guanidium, imidazole (Arg, Lys, His)

Zwitterion: form where some groups are charged but overall charge of molecule is 0

\[
\text{Glycine (and many others) at physiological pH}
\]

\[
\text{pK}_a = \text{pH for a given functional group where half of all molecules are protonated}
\]

\[
\begin{align*}
\text{pI} & = \text{pH where entire molecule has net charge of 0} \\
& \quad \text{If AA has 2 } pK_a \text{ values } \rightarrow \text{average them!} \\
& \quad \text{If AA has 3 } pK_a \text{ values, average most relevant (2 acidic or 2 basic)}
\end{align*}
\]
Amino Acids

15. Which type of chromatography would most effectively separate Ala from other amino acids with dissimilar properties in a sample at pH 2.7?

A) Cation-exchange chromatography  
B) Anion-exchange chromatography  
C) Size-exchange chromatography  
D) Nickel affinity chromatography

16. Of the following amino acids, the one most likely to be a neutral zwitterion is:

A) alanine at pH 5.  
B) tyrosine at pH 12.  
C) lysine at pH 4.  
D) glycine at pH 1.
17. In chemical hair-straightening, curly hair is relaxed into straight hair via alkalis used to reduce the disulfide bonds formed between residues in keratin proteins. What is the most likely target and function of this alkali treatment?

A) Threonine; the alkalis reduce, and thus form, new disulfide bonds between threonine residues.
B) Serine; the alkalis reduce, and thus form, new disulfide bonds between serine residues.
C) Cysteine; the alkalis reduce, and thus break, existing disulfide bonds between cysteine residues.
D) Methionine; the alkalis reduce, and thus break, existing disulfide bonds between methionine residues.

18. A professor attempts to purify glycine from a solution using anion-exchange chromatography. At which pH would he observe the largest amount of glycine adhering to the column?

A) 1.5
B) 6.5
C) 8.0
D) 10.0
Amino Acids

19. Due to the planar properties of the peptide bond, proteins can easily assume various organized structures (for example, beta sheets). All of the following are characteristics of the peptide bond EXCEPT:

A) its rotation is restricted.
B) it has multiple resonance forms.
C) it frequently breaks and reforms to allow structural fluidity.
D) it exhibits partial double bond character.

20. Ion-exchange chromatography with a positively-charged stationary phase is used to separate two polypeptides. Which amino acid is LEAST likely to be found in the polypeptide that elutes first?

A) A  B) H  C) P  D) D
21. Which of the following amino acid sequences would incur the greatest entropic penalty if it were used to replace Tyr-Cys-Met in the surface region of a protein?

A) His-Gly-Gly
B) Ala-Gly-Ser
C) Leu-Val-Phe
D) Met-Thr-Glu

22. At pH = 1, what is the net electrical charge on Tyr?

A) -1
B) 0
C) +1
D) +2
Amino Acids

23. Which substitution, of those below, is most likely to cause a change in the tertiary structure of a protein?

A) Val to Met
B) Lys to Leu
C) Ser to Thr
D) Asp to Glu

24. What structural characteristic marks the side chain of the amino acid N?

A) A four-carbon chain attached to an amine that makes the residue basic overall
B) A sulfur atom in the form of a thioether
C) A simple one-carbon group
D) An amide
Difficult Passage

The artificial sweetener aspartame is the methyl ester of the dipeptide of L-phenylalanine and L-aspartic acid (Figure 1). There are two general approaches to prepare aspartame. The chemical approach involves reacting the methyl ester of phenylalanine with an N-protected anhydride of aspartic acid. The protecting group, either a benzyl or formyl group is then removed by mild acid hydrolysis. In addition to the desired product, a beta structural isomer is also formed due to formation of a peptide bond with the wrong carboxylate group, which must be removed since it produces a bitter taste. A second enzymatic synthesis has been developed in which proteases catalyze the selective peptide bond formation and avoids the formation of the beta isomer.

Figure 1 Structure of N-(L-α-Aspartyl)-L-phenylalanine, 1-methyl ester (Note: $\text{pK}_{a1} = 3.2$ and $\text{pK}_{a2} = 7.7$)
Upon ingestion, aspartame is broken down in the duodenum into its components, aspartic acid, phenylalanine and methanol, with the subsequent formation of metabolites such as formaldehyde and formic acid. Some research has raised concerns that aspartame may lead to the formation of certain cancers as a result of the formation of some of these potentially toxic compounds. A new drug, known as protein AT7 (MW = $5 \times 10^4$ amu), has been developed to counter this possibility.
25. According to the passage, the pI of aspartame is most nearly:

A. 3.2  
B. 5.5  
C. 7.0  
D. 7.7

26. How many stereocenters are in aspartame?

A. 1  
B. 2  
C. 3  
D. 4
27. The two amino acids that form the basis for the dipeptide structure of aspartame, aspartic acid and phenylalanine, are most accurately be classified as:

A. hydrophilic and hydrophilic, respectively.
B. hydrophobic and hydrophilic, respectively.
C. hydrophilic and hydrophobic, respectively.
D. hydrophobic and hydrophobic, respectively.

28. Prior to its digestion in the small intestine, aspartame must pass through the stomach. What is net charge on aspartame while in the stomach?

A. -1
B. 0
C. +1
D. +2
29. How many amino acid residues are in AT7?

A. 2  
B. 50  
C. 450  
D. 900

30. Peptides are stable in water because:

A) peptide bonds cannot be cleaved by hydrolysis.  
B) electron sharing between the carbonyl and amino group contributes resonance stabilization across the amide bond.  
C) the breakdown of peptides into individual amino acids is entropically unfavorable.  
D) peptides hydrogen bond with free-floating proline residues to promote stabilization.
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Custom study plan to show you your path

1. Select your start date
   - 08/10/2016
2. Select your exam date
   - 10/31/2016

3. How will you be preparing for the MCAT?
   - On the side (5-20 hrs/week)
   - Part-Time (20-40 hrs/week)
   - Full-Time (40+ hrs/week)

4. Select the MCAT science subject about which you are most worried
   - Biology
   - Biochem
   - General Chemistry
   - Organic Chemistry
   - Physics
   - Psy/Soc

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### Scaled Scores

<table>
<thead>
<tr>
<th>Section</th>
<th>Scaled Scores</th>
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### Results by Reasoning Skills

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<th>Concept Category</th>
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<th>Correct Percentage</th>
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<td>49</td>
<td>71.01% (49/69)</td>
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<tr>
<td>2 Scientific Reasoning and Problem Solving</td>
<td>39</td>
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<td>3 Reasoning About the Design and Execution of Research</td>
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<td>4 Data-based and Statistical Reasoning</td>
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### Results by Section

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Princeton

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Kaplan

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