Chapter 5: Microbial Metabolism

1. Enzymes

2. ATP Production

3. Autotrophic Processes
1. **Enzymes**

Chapter Reading – pp. 126-134
Biochemical Reactions

All living cells depend on biochemical reactions to maintain homeostasis.

All of the biochemical reactions in an organism are collectively referred to as metabolism, which is of 2 basic types:

- **catabolic:** reactions that “break down” molecules
  - generally energy releasing or **exergonic**

- **anabolic:** reactions that build new molecules
  - generally energy requiring or **endergonic**

**exergonic reactions provide energy for endergonic ones!**
All organisms, prokaryotic or eukaryotic, need to build the molecules they need, and find the energy to do so!
Metabolic Pathways

Most biochemical reactions are part of a series of reactions referred to as a metabolic pathway:

- it usu. takes multiple reactions to make “end-product”
- pathways can be catabolic or anabolic
- each reaction is catalyzed by its own enzyme
Enzyme Basics

Almost all biochemical reactions are catalyzed by a specific enzyme:

- proteins that accelerate the rate of a reaction without being changed themselves
  - lower the activation energy ($E_a$)

- the need for enzymes provides a way to control or regulate biochemical reactions
  - reactions won’t occur unless the enzyme that catalyzes the reaction is present & active
Enzymes lower the Activation Energy

**Reactions won't occur unless the \( E_a \) requirement is met**
Enzymes physically bind Substrates

The “fit” of substrate into active site is highly specific and due to molecular complementarity

- complementary in physical shape ("hand in glove")
- complementary in chemical properties (attraction between opposite charges, hydrophobic regions)
Control of Enzyme Activity

Biochemical reactions can be controlled by changes in enzyme activity, which can be influenced in several ways:

1) Changes in the amount of enzyme or substrate
   - more enzyme &/or more substrate = more product!

2) Changes in temperature, pH or [salt]
   - can effect enzyme structure, hence its activity

3) Availability of any necessary cofactors
   - some enzymes don’t work w/o a non-protein cofactor

4) Effect of inhibitors
   - molecules that bind to enzymes & reduce their activity
Factors effecting Enzyme Activity

Reactions occur more rapidly as temperature rises, until temperature is too high and the enzyme is denatured.

Enzyme structure depends on pH
- pH affects R group interactions and, hence, protein structure
- pH extremes denature enzymes

Reaction rates increase with higher substrate concentrations, then level off when enzyme is saturated.
Enzyme Denaturation

Enzymes are polypeptides that retain their ability to function only when folded properly.

- changes in pH temperature, or [salt] can disrupt amino acid “R group” interactions causing the protein to misfold, i.e., become denatured

**mutations can also lead to misfolded, non-functional enzymes**
Some Enzymes Require Cofactors

- can be a metal ion, vitamin, or other “non-protein”
  - if the cofactor is organic, it is called a coenzyme
- enzyme is inactive without cofactor
Competitive Enzyme Inhibition

Competitive inhibition involves binding of an inhibitor to the active site.

- inhibitor must be reversible to be able to regulate in response to concentration

Irreversible inhibitors essentially poison the enzyme.
Allosteric Enzyme Regulation

Allosteric regulation involves the binding of a substance to an enzyme outside the active site.

- Induces change in shape of active site
- Must be reversible

(a) Allosteric inhibition

(b) Allosteric activation
Feedback Inhibition

The end-products of metabolic pathways are important reversible enzyme inhibitors.

- inhibit 1st enzyme in pathway, turning the pathway “off”
  - low [inhibitor] = pathway ON
  - high [inhibitor] = pathway OFF

- can be competitive or allosteric inhibition

- provide an important way of regulating end-product levels
2. ATP Production

Chapter Reading – pp. 134-148
Adenosine Triphosphate (ATP)

Preferred source of useable energy for ALL cells:

- breaking bond of 3rd phosphate releases ideal amt of energy
- bond is easily broken (low $E_a$)

**This is why organisms convert “food” energy to “ATP” energy**
3 General Types of ATP Production

**Substrate-Level Phosphorylation**
- direct transfer of $P$ from organic molecule to $\text{ADP} \rightarrow \text{ATP}$

**Oxidative Phosphorylation**
- oxidation/reduction reactions provide energy for $\text{ADP} + P \rightarrow \text{ATP}$

**Photophosphorylation**
- light provides energy for $\text{ADP} + P \rightarrow \text{ATP}$
How is ATP produced?

In most organisms, energy from a “food source” is converted to energy in ATP by **glycolysis** followed by 1 of 2 processes:

**FERMENTATION** (low ATP yield)

or

**RESPIRATION** (high ATP yield)
Glycolysis

Glycolysis is a catabolic pathway by which sugars such as glucose (& several other “food” sources) are broken down to two 3-Carbon molecules of pyruvic acid (or pyruvate):

- releases energy to yield 2 ATP per glucose
- also transfers high energy electrons (+ H) to NAD$^+$ to yield 2 NADH
Oxidation/Reduction

Much of the energy in “food” molecules such as glucose is captured as high energy electrons (e^-) by electron carriers such as NADH & FADH$_2$

- When a molecule receives or gains electrons it is said to be reduced
  - e^- are typically transferred as part of a Hydrogen atom

- A molecule that gives up electrons (i.e., loses H) is said to be oxidized
Fermentation

ATP production begins & ends with glycolysis in organisms that ferment.

Fermentation is all about regenerating NAD\(^+\) so that glycolysis can continue:

- NADH is oxidized to NAD\(^+\) by reducing pyruvate to lactic acid for example.
The Variety of Fermentation Products

Glucose $\rightarrow$ NAD$^+$ $\rightarrow$ NADH $\rightarrow$ Pyruvate

Propionibacterium: CO$_2$, propionic acid (Swiss cheese)
Aspergillus Lactobacillus Streptococcus: Lactic acid (Cheddar cheese, yogurt, soy sauce)
Saccharomyces: CO$_2$, ethanol (Wine, beer)
Clostridium: Acetone, isopropanol (Nail polish remover, rubbing alcohol)
Escherichia Acetobacter: Acetic acid (Vinegar)
Respiration

After glycolysis, energy in pyruvate & NADH is used to produce much more ATP by respiration:

**KREBS CYCLE**
- breaks down pyruvate to 3 CO₂, energy captured as e⁻ by NADH & FADH₂

**ELECTRON TRANSPORT**
- e⁻ from NADH, FADH₂ used to produce H⁺ gradient

**CHEMIOSMOSIS**
- H⁺ gradient used to make ATP
The Krebs cycle

- a cyclical metabolic pathway catalyzed by enzymes in the matrix of mitochondria
- requires 2-C acetyl groups connected to coenzyme A (acetyl-CoA)

\[
\text{(3-C) pyruvate} + \text{CoA} \\
\text{(2-C) acetyl-CoA} + \text{CO}_2
\]

(Krebs cycle)
Electron Transport & Chemiosmosis

Occurs in the mitochondria of eukaryotes and at the plasma membrane of prokaryotes.

- oxygen \((O_2)\) is usually the final electron acceptor, but other molecules can play this role in \textit{anaerobic} respiration.
Lipid Catabolism

(a) Hydrolysis
- Free fatty acids are released from triglycerides

(b) Beta-oxidation
- Fatty acids converted to Acetyl-CoA units
  - To electron transport chain
  - To Krebs cycle

a. Hydrolysis – free fatty acids are released from triglycerides
b. Beta Oxidation – fatty acids converted to Acetyl-CoA units
Protein Catabolism

Extracellular fluid

Proteases

Polypeptide

H₂O

A

Amino acids

Cytoplasmic membrane

Cytoplasm

Deamination

H₂O

NH₂

2H

B

C—C—OH → To Krebs cycle
Lipid & Protein Catabolism

Lipids and proteins can also be used as sources of energy to produce ATP

- different amino acids enter glycolysis or the Krebs cycle at various stages
- fatty acids are broken down to acetyl groups & fed into the Krebs cycle
Summary of ATP Production

<table>
<thead>
<tr>
<th>Energy-Producing Process</th>
<th>Growth Conditions</th>
<th>Final Hydrogen (Electron) Acceptor</th>
<th>Type of Phosphorylation Used to Generate ATP</th>
<th>ATP Molecules Produced per Glucose Molecule</th>
</tr>
</thead>
<tbody>
<tr>
<td>Aerobic Respiration</td>
<td>Aerobic</td>
<td>Molecular oxygen (O_2)</td>
<td>Substrate-level and oxidative</td>
<td>36 (eukaryotes) 38 (prokaryotes)</td>
</tr>
<tr>
<td>Anaerobic Respiration</td>
<td>Anaerobic</td>
<td>Usually an inorganic substance (such as (NO_3^-), (SO_4^{2-}), or (CO_3^{2-}) but not molecular oxygen (O_2))</td>
<td>Substrate-level and oxidative</td>
<td>Variable (fewer than 38 but more than 2)</td>
</tr>
<tr>
<td>Fermentation</td>
<td>Aerobic or anaerobic</td>
<td>An organic molecule</td>
<td>Substrate-level</td>
<td>2</td>
</tr>
</tbody>
</table>

**Obligate anaerobes:**
- fermentation or anaerobic respiration

**Obligate aerobes:**
- aerobic respiration (& brief periods of fermentation)

**Facultative anaerobes:**
- can survive via aerobic respiration OR fermentation
Key Terms for Chapter 5

• catabolic, anabolic; exergonic, endergonic
• activation energy, substrate, active site
• cofactor vs coenzyme, denatured
• feedback inhibition: competitive vs allosteric
• glycolysis, fermentation, respiration
• Krebs cycle, electron transport, chemiosmosis
• oxidation vs reduction
• substrate-level, oxidative & photophosphorylation

Relevant Chapter Questions
MC: 1-11, 13-16, 19-20    M: 1-4    FI: 2, 4, 5, 7