<u>Chapter 5:</u> <u>Microbial Metabolism</u>

1. Enzymes

2. ATP Production

3. Autotrophic Processes

1. Enzymes

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 <u>metabolism</u>, which is of 2 basic types:

catabolic: reactions that "break down" molecules

• generally energy *releasing* or <u>exergonic</u>

anabolic: reactions that build new molecules

• generally energy *requiring* or <u>endergonic</u>

***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 <u>metabolic pathway</u>:

- it usu. takes multiple reactions to make "end-product"
- pathways can be <u>catabolic</u> or <u>anabolic</u>
- each reaction is catalyzed by its own enzyme

Enzyme Basics

Almost all biochemical reactions are *catalyzed* by a specific <u>enzyme</u>:

- 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 <u>& active</u>

Enzymes lower the Activation Energy



Enzymes physically bind Substrates



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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







Temperature

- reactions occur more rapidly as temperature rises
- **as long as enzyme is active (heat can <u>denature</u> enzymes)

рΗ

 enzyme structure depends on pH

**pH affects charge
 of "R groups",
 protein structure

[Substrate]

- reactions occur more rapidly as [substrate] rises
- **<u>saturation</u> occurs when [substrate] is high enough

Enzyme Denaturation

- enzymes are polypeptides that retain their ability to function only when folded properly
- changes in temperature, pH or [salt] can disrupt amino acid "R group" interactions causing the protein to unfold, i.e. become <u>denatured</u>



Active (functional) protein Denatured protein
mutations can also lead to misfolded, non-functional enzymes

Some Enzymes Require Cofactors



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can be a metal ion, vitamin, or other "non-protein"

- if the cofactor is organic, it is called a coenzyme
- enzyme is inactive w/o cofactor

Enzyme Inhibition



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- inhibitors bind enzymes in 1 of 2 ways:
 - competitive inhibition (binding to active site)
 - allosteric inhibition (binding elsewhere, changing shape)
- inhibitors can bind <u>reversibly</u> (can "come off") or <u>irreversibly</u> (don't come off, e.g. "poisons")



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Feedback Inhibition

The end-products of metabolic pathways are important <u>reversible</u> 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

Adenosine Triphosphate (ATP)



Preferred source of <u>useable</u> 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

(active



How is ATP produced? In most organisms,

In most organisms, energy from a "food source" is converted to energy in ATP by <u>glycolysis</u> followed by 1 of 2 processes:

> FERMENTATION (low ATP yield)

> > or

RESPIRATION (high ATP yield)

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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 & FADH₂



 when a molecule receives or gains electrons it is said to be <u>reduced</u>

> **e⁻ are typically transferred as part of a Hydrogen atom**

> > • a molecule that gives up electrons (i.e., loses H) is said to be <u>oxidized</u>

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Fermentation

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

Fermentation is all about recycling NAD⁺ so that glycolysis can continue:







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Different Fermentation Products

TABLE 5.4 Som	e Industrial Uses for Different Types Of Fermentations			
Fermentation End-Product(s)	Industrial or Commercial Use	Starting Material	Microorganism	
Ethanol	Beer	Malt extract	Saccharomyces cerevisiae (yeast, a fungus)	
	Wine	Grape or other fruit juices	Saccharomyces cerevisiae var. ellipsoideus	
	Fuel	Agricultural wastes	Saccharomyces cerevisiae	
Acetic Acid	Vinegar	Ethanol	Acetobacter (bacterium)	
Lactic Acid	Cheese, yogurt	Milk	Lactobacillus, Streptococcus (bacteria)	
	Rye bread	Grain, sugar	Lactobacillus delbruckii (bacterium)	
	Sauerkraut	Cabbage	Lactobacillus plantarum (bacterium)	
	Summer sausage	Meat	Pediococcus (bacterium)	
Propionic Acid and Carbon Dioxide	Swiss cheese	Lactic acid	Propionibacterium freudenreichii (bacterium)	
Acetone and Butanol	Pharmaceutical, industrial uses	Molasses	Clostridium acetobutylicum (bacterium)	
Glycerol	Pharmaceutical, industrial uses	Molasses	Saccharomyces cerevisiae	
Citric Acid	Flavoring	Molasses	Aspergillus (fungus)	
Methane	Fuel	Acetic acid	Methanosarcina (bacterium)	
Sorbose	Vitamin C (ascorbic acid)	Sorbitol	Gluconobacter	

Different organisms recycle NAD⁺ in different ways, resulting in a variety of fermentation end-products.



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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 <u>acetyl</u> groups connected to coenzyme A (acetyl-CoA)

(3-C) pyruvate + CoA \downarrow (2-C) acetyl-CoA + CO₂ \vdots (Krebs cycle)

Electron Transport & Chemiosmosis



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

 oxygen (O₂) is usually the <u>final electron acceptor</u>, but other molecules can play this role in *anaerobic* respiration



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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 5.5	Aerobic Respiration, Anaerobic Respiration, and Fermentation Compared				
Energy-Producing Process	Growth Conditions	Final Hydrogen (Electron) Acceptor	Type of Phosphorylation Used to Generate ATP	ATP Molecules Produced per Glucose Molecule	
Aerobic Respiration	Aerobic	Molecular oxygen (O ₂)	Substrate-level and oxidative	36(eukaryotes) 38 (prokaryotes)	
Anaerobic Respiration	Anaerobic	Usually an inorganic substance (such as NO_3^- , SO_4^{2-} , or CO_3^{2-}) but not mo- lecular oxygen (O ₂)	Substrate-level and oxidative	Variable (fewer than 38 but more than 2)	
Fermentation	Aerobic or anaerobic	An organic molecule	Substrate-level	2	

Obligate anaerobes:

fermentation or anaerobic respiration

Obligate aerobes:

aerobic respiration (& brief periods of fermentation)

Facultative anaerobes:

• can survive via aerobic respiration OR fermentation

3. <u>Autotrophic Processes</u>

All organisms depend on Autotrophs

Autotrophs can produce organic molecules from CO₂, an inorganic carbon source.

- all heterotrophs require an organic source of carbon
- organic molecules, directly or indirectly, come from autotrophs
- The source of energy for autotrophic processes can be:
 - LIGHT: photoautotrophs that carry out photosynthesis

CHEMICAL: chemoautotrophs that use various molecules as a source of high energy e⁻

Light Reactions of Photosynthesis

Electrons (from H₂O) energized by sunlight:

 fuel the synthesis of ATP through electron transport & chemiosmosis (much like respiration)





"Dark" Reactions

Involves an anabolic pathway known as the Calvin-Benson cycle:

 endergonic reactions of this pathway are fueled by ATP & NADPH from the "light" reactions

 resulting sugars can be used as a source of energy or to build other organic molecules

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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
- Calvin-Benson cycle

Relevant Chapter Questions rvw: 1-7, 18, 20-22 MC: 1, 4-10