Chapter 5  
Microbial Nutrition

**Movies:**
1) Active transport by group translocation
2) Cotransport (symport and antiport)
3) How diffusion work
4) How osmosis work
5) Limiting amino acids

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The Common Nutrient Requirements

- **Macroelements (macronutrients)**
  - C, O, H, N, S, P, K, Ca, Mg, and Fe
  - required in relatively large amounts

- **Micronutrients (trace elements)**
  - Mn, Zn, Co, Mo, Ni, and Cu
  - required in trace amounts
  - often supplied in water or in media components

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All Organisms Require Carbon, Hydrogen, Oxygen and Electron Source

- Carbon is backbone of all organic components present in cell
- Hydrogen and oxygen are also found in organic molecules
- Electrons play a role in energy production and reduction of CO₂ to form organic molecules

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Requirements for Carbon, Hydrogen, and Oxygen

- Often satisfied together
  - Carbon source often provides H, O, and electrons
- **Heterotrophs**
  - Use organic molecules as carbon sources which often also serve as energy source
- **Autotrophs**
  - Use carbon dioxide as their sole or principal carbon source
  - Must obtain energy from other sources

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Sources of Carbon, Energy and Electrons

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<th>Table 5.1</th>
<th>Sources of Carbon, Energy, and Electrons</th>
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<tr>
<td><strong>Carbon Sources</strong></td>
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<tr>
<td>Autotrophs</td>
<td>CO₂ sole or principal biosynthetic carbon source (section 10.3)</td>
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<tr>
<td>Heterotrophs</td>
<td>Reduced, preformed, organic molecules from other organisms (chapters 9 and 10)</td>
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<tr>
<td><strong>Energy Sources</strong></td>
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<tr>
<td>Phototrophs</td>
<td>Light (section 9.12)</td>
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<td>Chemotrophs</td>
<td>Oxidation of organic or inorganic compounds (chapter 9)</td>
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<td><strong>Electron Sources</strong></td>
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<tr>
<td>Lithotrophs</td>
<td>Reduced inorganic molecules (section 9.11)</td>
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<tr>
<td>Organotrophs</td>
<td>Organic molecules (chapter 9)</td>
</tr>
</tbody>
</table>

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Nutritional Types of Organisms

- **Based on energy source**
  - Phototrophs use light
  - Chemotrophs obtain energy from oxidation of chemical compounds
- **Based on electron source**
  - Lithotrophs use reduced inorganic substances
  - Organotrophs obtain electrons from organic compounds
Nutritional Types of Microorganisms

<table>
<thead>
<tr>
<th>Nutritional Type</th>
<th>Carbon Source</th>
<th>Energy Source</th>
<th>Electron Source</th>
<th>Representative Microorganisms</th>
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</thead>
<tbody>
<tr>
<td>Phototrophs</td>
<td>Organic</td>
<td>Light</td>
<td>Organic</td>
<td>Photosynthetic bacteria, cyanobacteria</td>
</tr>
<tr>
<td>Chemotrophs (photoheterotrophs)</td>
<td>Organic</td>
<td>Inorganic</td>
<td>Organic</td>
<td>Putrid bacteria, sulfate reducers</td>
</tr>
</tbody>
</table>

Requirements for Nitrogen, Phosphorus, and Sulfur

- Needed for synthesis of important molecules (e.g., Amino acids, nucleic acids)
- Nitrogen supplied in numerous ways
- Phosphorus usually supplied as inorganic phosphate
- Sulfur usually supplied as sulfate via assimilatory sulfate reduction

Sources of nitrogen

- Organic molecules
- Ammonia
- Nitrate via assimilatory nitrate reduction
- Nitrogen gas via nitrogen fixation

Sources of Phosphorus and Sulfur

- Phosphorus
  - Most organisms use inorganic phosphorus which is directly incorporated into their cells
- Sulfur
  - Most organisms use sulfate and reduce it by assimilatory sulfate reduction

Growth Factors

- Organic compounds
- Essential cell components (or their precursors) that the cell cannot synthesize
- Must be supplied by environment if cell is to survive and reproduce

Classes of growth factors

- Amino acids
  - Needed for protein synthesis
- Purines and pyrimidines
  - Needed for nucleic acid synthesis
- Vitamins
  - Function as enzyme cofactors
Uptake of Nutrients by the Cell

- Some nutrients enter by passive diffusion
- Most nutrients enter by:
  - Facilitated diffusion
  - Active transport
  - Group translocation

Movies
Active transport by group translocation
Cotransport (symport and antiport)
How diffusion work

Passive Diffusion
- Molecules move from region of higher concentration to one of lower concentration because of random thermal agitation
- \( \text{H}_2\text{O}, \text{O}_2 \) and \( \text{CO}_2 \) often move across membranes this way

Facilitated Diffusion
- Similar to passive diffusion
  - Movement of molecules is **not** energy dependent
  - Direction of movement is from high concentration to low concentration
  - Size of concentration gradient impacts rate of uptake

Facilitated diffusion...
- Differs from passive diffusion
  - Uses carrier molecules (permeases)
  - Smaller concentration gradient is required for significant uptake of molecules
  - Effectively transports glycerol, sugars, and amino acids
  - More prominent in eucaryotic cells than in procaryotic cells

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<table>
<thead>
<tr>
<th>Vitamin</th>
<th>Function</th>
<th>Examples of Vitaminostats Involveda</th>
</tr>
</thead>
</table>

| B (thiamin) | Cofactor for many enzymes; coenzyme B1 (thiamin pyrophosphate) | Pyruvate dehydrogenase, \( \text{H}_2\text{O} \), \( \text{O}_2 \), \( \text{CO}_2 \) often move across membranes this way |
| Vitamin C (ascorbic acid) | Reduces oxidized substances; coenzyme \( \text{H}^+ \) | \( \text{H}^+ \) carriers, \( \text{H}^+ \) gradients, \( \text{H}^+ \) ATPase |
| Folate | ONE-carbon transfers; coenzyme F | \( \text{H}^+ \) pumps, \( \text{H}^+ \) gradients, \( \text{H}^+ \) ATPase |
| I (iodine) | Transfer of iodine ions; coenzyme I | \( \text{H}^+ \) pumps, \( \text{H}^+ \) gradients, \( \text{H}^+ \) ATPase |
| Niacin (niacinamide) | Pyridine nucleotides; coenzyme NAD, NADP | \( \text{H}^+ \) pumps, \( \text{H}^+ \) gradients, \( \text{H}^+ \) ATPase |
| Pantothenic acid | Coenzyme A; precursor to cAMP, \( \text{H}^+ \) gradients, \( \text{H}^+ \) ATPase |
| Riboflavin (vitamin B2) | Flavin nucleotides; coenzyme FAD, FADH \(_2\) | \( \text{H}^+ \) pumps, \( \text{H}^+ \) gradients, \( \text{H}^+ \) ATPase |
| Thiamin (vitamin B1) | Pyruvate dehydrogenase complex; coenzyme B1 (thiamin pyrophosphate) | \( \text{H}^+ \) pumps, \( \text{H}^+ \) gradients, \( \text{H}^+ \) ATPase |
| Vitamin B6 (pyridoxine) | Pyridoxal phosphate; coenzymes | \( \text{H}^+ \) pumps, \( \text{H}^+ \) gradients, \( \text{H}^+ \) ATPase |

\( a \) The enzymes involved are numbered and function: kinase (k), hydrolase (h), ligase (l), oxidoreductase (o), and transferase (t).
**Active Transport**

- Energy-dependent process
  - ATP or proton motive force used
- Moves molecules against the gradient
- Concentrates molecules inside cell
- Involves carrier proteins (permeases)
  - Carrier saturation effect is observed at high solute concentrations

**ABC transporters**

- ATP-binding cassette transporters
- Observed in bacteria, archaea, and eucaryotes

**Active transport using proton and sodium gradients**

**Group Translocation**

- Chemically modifies molecule as it is brought into cell
- Best known system: transports a variety of phosphoenolpyruvate: sugar phosphotransferase system (PTS)
  - Sugars while phosphorylating them using phosphoenolpyruvate (PEP) as the phosphate donor
Iron Uptake

- Ferric iron is very insoluble so uptake is difficult
- Microorganisms use siderophores to aid uptake
- Siderophore complexes with ferric ion
- Complex is then transported into cell

![Siderophore ferric iron complexes](image)

Culture Media

- Most contain all the nutrients required by the organism for growth
- Classification
  - Chemical constituents from which they are made
  - Physical nature
  - Function

Types of Media

<table>
<thead>
<tr>
<th>Physical Nature</th>
<th>Chemical Composition</th>
<th>Functional Type</th>
</tr>
</thead>
<tbody>
<tr>
<td>Liquid</td>
<td>Defined (synthetic)</td>
<td>Supportive (general purpose)</td>
</tr>
<tr>
<td>Semisolid</td>
<td>Complex</td>
<td>Enriched</td>
</tr>
<tr>
<td>Solid</td>
<td></td>
<td>Selective</td>
</tr>
<tr>
<td></td>
<td></td>
<td>Differential</td>
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</tbody>
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Defined or Synthetic Media

- All components and their concentrations are known

Some media components

- Peptones
  - Protein hydrolysates prepared by partial digestion of various protein sources
- Extracts
  - Aqueous extracts, usually of beef or yeast
- Agar
  - Sulfated polysaccharide used to solidify liquid media
**Functional Types of Media**

- **Supportive or general purpose media**
  - Support the growth of many microorganisms
  - E.g., Tryptic soy agar
- **Enriched media**
  - General purpose media supplemented by blood or other special nutrients
  - E.g., Blood agar

**Types of media**

- **Selective media**
  - Favor the growth of some microorganisms and inhibit growth of others
  - E.g., Macconkey agar
    - Selects for gram-negative bacteria

**Differential media**

- Distinguish between different groups of microorganisms based on their biological characteristics
  - E.g., blood agar
    - Hemolytic versus nonhemolytic bacteria
  - E.g., MacConkey agar
    - Lactose fermenters versus nonfermenters

**Isolation of Pure Cultures**

- **Pure culture**
  - Population of cells arising from a single cell
- **Spread plate, streak plate, and pour plate**
  - Techniques used to isolate pure cultures
The Spread Plate and Streak Plate

- Involve spreading a mixture of cells on an agar surface so that individual cells are well separated from each other
- Each cell can reproduce to form a separate colony (visible growth or cluster of microorganisms)

Spread-plate technique

1. dispense cells onto medium in petri dish
2. - 3. sterilize spreader
4. spread cells across surface

Appearance of a Spread Plate

Streak plate technique

Note: This method only works if the spreading test plate or inoculating loop is sterilized after each streak. OD.

The Pour Plate

- Sample is diluted several times
- Diluted samples are mixed with liquid agar
- Mixture of cells and agar are poured into sterile culture dishes
Colony growth

- Most rapid at edge of colony
  - Oxygen and nutrients are more available at edge
- Slowest at center of colony
- In nature, many microorganisms form biofilms on surfaces