Microbial Metabolism

By,

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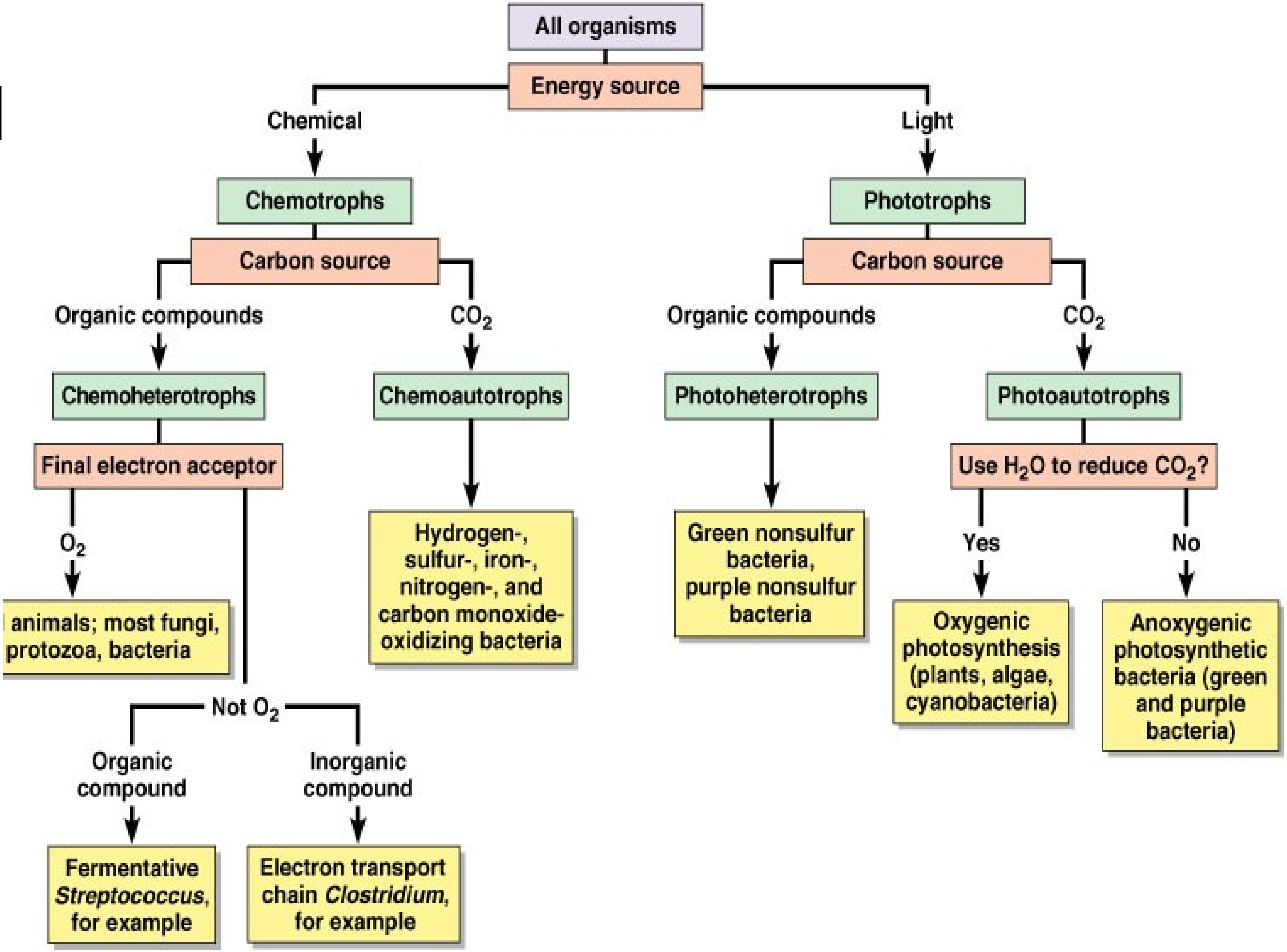
Synopsis:

A. Basic Concepts of Metabolism
B. Glycolytic Pathways
C. Fermentation
D. Respiration
E. Photosynthesis
F. Chemolithotrophy
G. The Nitrogen Cycle

INTRODUCTION

Bacterial growth = ↑ size and number

1. **Basic requirement = C, H, O, N, inorganic salts.**
2. **AUTOTROPHS = Bacteria which can synthesize their own food.**
3. **HETEROTROPHS = bacteria which cannot synthesize their own food.**


All organisms
Energy source
Chemical
Light
Chemotrophs
Phototrophs
Carbon source
Carbon source
Organic compounds
CO_2
Organic compounds
CO_2
Chemoheterotrophs
Chemoautotrophs
Photoheterotrophs
Photoautotrophs
Final electron acceptor
Use H_2O to reduce CO_2?
O_2
animals; most fungi, protozoa, bacteria
Not O_2
Organic compound
Inorganic compound
Fermentative *Streptococcus*, for example
Electron transport chain *Clostridium*, for example
Hydrogen-, sulfur-, iron-, nitrogen-, and carbon monoxide-oxidizing bacteria
Green nonsulfur bacteria, purple nonsulfur bacteria
Yes
No
Oxygenic photosynthesis (plants, algae, cyanobacteria)
Anoxygenic photosynthetic bacteria (green and purple bacteria)

NUTRITIONAL REQUIREMENTS

1. **Essential elements – C,H,O,N, phosphorous, sulphur.**

2. **Mineral sources – Na, K, Mg, Fe, Mn, Ca, Co etc.**

3. **Organic growth factors**

NUTRITIONAL REQUIREMENTS

- **ESSENTIAL ELEMENTS:**

- **Bacterial structure is made up of various components eg. carbohydrates, lipids, proteins, nucleic acids.**

- **These compounds are made up of four basic elements viz. C, H, O, and N.**

- **Besides these four elements, phosphorous and sulphur are also required for bacterial growth.**

a. **Hydrogen and oxygen** = supplied from water added to the culture medium.

b. **Carbon** = supplied by carbohydrates. Bacteria also get energy in the form of ATP from carbohydrate breakdown.

c. **Nitrogen** = it is a major component of proteins and nucleic acids. It is obtained from mainly ammonia, usually in the form of ammonium salts. Ammonium salts are obtained from environment or from deamination of amino acids.

d. Sulphur = It is a part of proteins and coenzymes. Sulphur is obtained from sulphates. Many bacteria reduce these sulphates to hydrogen sulphide (H_2S).

e. Phosphorous = it is required for nucleic acids, ATP, coenzymes etc.

2. MINERALS:

1. Potassium, calcium, magnesium, iron, copper, cobalt, manganese, molybdenum and zinc.

2. These elements are required in trace amounts and are provided from various food sources.

3. ORGANIC GROWTH FACTORS

1. Organic compounds are required by bacteria for their growth and maintenance.

2. They are called BACTERIAL VITAMINS.

3. Bacteria have a very variable growth requirement.

4. A growth factor may be essential for some, and accessory for other bacteria, and totally unrequired by some others.

5. Eg. H.influenza requires accessory growth factors X & V.

A. Basic Concepts

1. Definitions

a) Metabolism: The processes of catabolism and anabolism

b) Catabolism: The processes by which a living organism obtains its energy and raw materials from nutrients

c) Anabolism: The processes by which energy and raw materials are used to build macromolecules and cellular structures (biosynthesis)

A. Basic Concepts

2. Reduction and Oxidation

a) An atom becomes more reduced when it undergoes a chemical reaction in which it

- Gains electrons
- By bonding to a less electronegative atom
- And often this occurs when the atom becomes bonded to a hydrogen

A. Basic Concepts

2. Reduction and Oxidation

a) An atom becomes more oxidized when it undergoes a chemical reaction in which it

- Loses electrons
- By bonding to a more electronegative atom
- And often this occurs when the atom becomes bonded to an oxygen

A. Basic Concepts

2. Reduction and Oxidation

c) In metabolic pathways, we are often concerned with the oxidation or reduction of carbon.

d) Reduced forms of carbon (e.g. hydrocarbons, methane, fats, carbohydrates, alcohols) carry a great deal of potential chemical energy stored in their bonds.

e) Oxidized forms of carbon (e.g. ketones, aldehydes, carboxylic acids, carbon dioxide) carry very little potential chemical energy in their bonds.

A. Basic Concepts

2. Reduction and Oxidation

f) Reduction and oxidation always occur together. In a reduction-oxidation reaction (redox reaction), one substance gets reduced, and another substance gets oxidized. The thing that gets oxidized is called the electron donor, and the thing that gets reduced is called the electron acceptor.

A. Basic Concepts

3. Enzymatic Pathways for Metabolism

a) Metabolic reactions take place in a step-wise fashion in which the atoms of the raw materials are rearranged, often one at a time, until the formation of the final product takes place.

b) Each step requires its own enzyme.

c) The sequence of enzymatically-catalyzed steps from a starting raw material to final end products is called an enzymatic pathway (or metabolic pathway)

A. Basic Concepts

4. Cofactors for Redox Reactions

a) Enzymes that catalyze redox reactions typically require a cofactor to “shuttle” electrons from one part of the metabolic pathway to another part.

b) There are two main redox cofactors: NAD and FAD. These are (relatively) small organic molecules in which part of the structure can either be reduced (e.g., accept a pair of electrons) or oxidized (e.g., donate a pair of electrons)

A. Basic Concepts

4. Cofactors for Redox Reactions

$NAD_{(oxidized)} + H^+ + \text{Pair of electrons} \rightarrow NADH_{(reduced)}$

$FAD_{(oxidized)} + H^+ + \text{Pair of electrons} \rightarrow FADH_{(reduced)}$

NAD and FAD are present only in small (catalytic) amounts – they cannot serve as the final electron acceptor, but must be regenerated (reoxidized) in order for metabolism to continue

A. Basic Concepts

5. **ATP: A "currency of energy" for many cellular reactions**

   a) **ATP stands for adenosine triphosphate. It is a nucleotide with three phosphate groups linked in a small chain.**

   b) **The last phosphate in the chain can be removed by hydrolysis (the ATP becomes ADP, or adenosine diphosphate).**
   This reaction is energetically favorable: it has a $\Delta G°'$ of about –7.5 kcal/mol

$$\text{ATP} + H_2O \rightarrow \text{ADP} + \text{Phosphate} + \text{Energy (7.5 kcal/mol)}$$

A. Basic Concepts

5. ATP

c) ATP hydrolysis is used as an energy source in many biological reactions that require energy – for example, active transport in the sodium-potassium pump

d) During catabolism, energy released from the oxidation of carbon is captured and used to synthesize ATP from ADP and phosphate.

$$C_6H_{12}O_6 + 6\ O_2 \rightarrow 6\ CO_2 + 6\ H_2O + \text{Energy}$$

$$\text{ADP} + \text{Phosphate} + \text{Energy} \rightarrow \text{ATP} + H_2O`$$

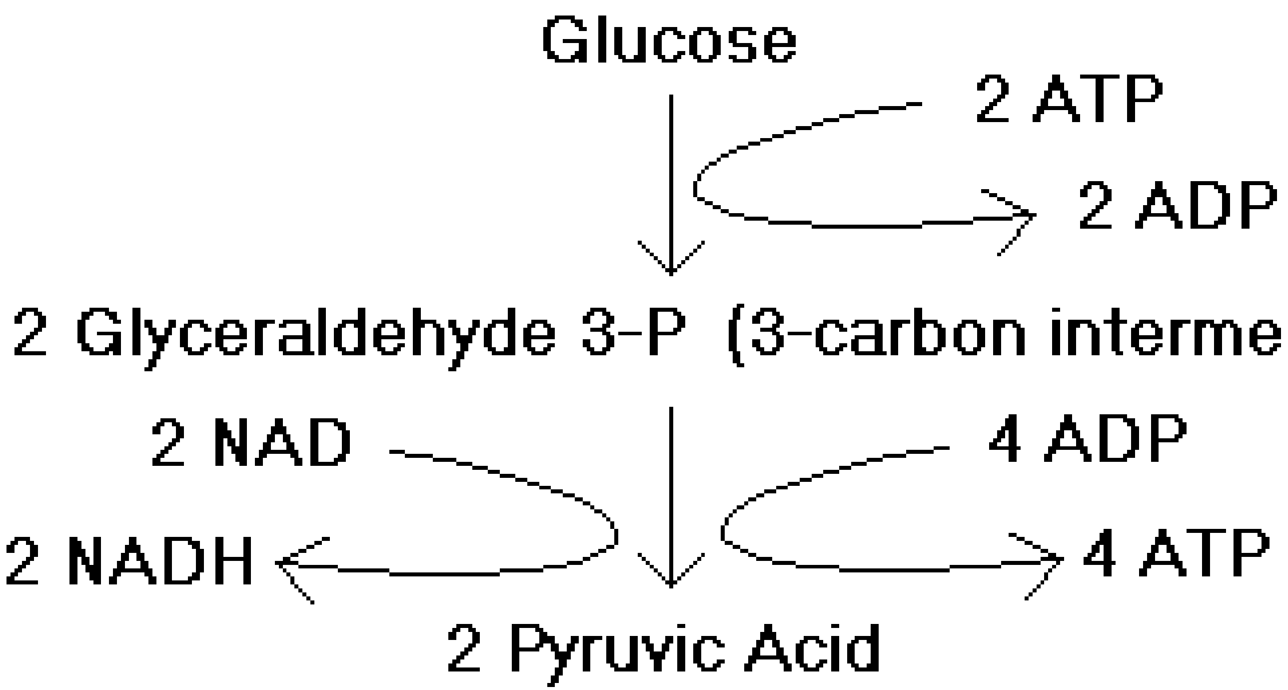
B. Glycolytic Pathways

1. Features of glycolytic pathways
   a) Partial oxidation of glucose to form pyruvic acid
   b) A small amount of ATP is made
   c) A small amount of NAD is reduced to NADH

B. Glycolytic Pathways

2. **4 major glycolytic pathways found in different bacteria:**
   a) **Embden-Meyerhoff-Parnas pathway**
      - **"Classic" glycolysis**
      - **Found in almost all organisms**
   a) **Hexose monophosphate pathway**
      - **Also found in most organisms**
      - **Responsible for synthesis of pentose sugars used in nucleotide synthesis**
   a) **Entner-Doudoroff pathway**
      - **Found in *Pseudomonas* and related genera**
   a) **Phosphoketolase pathway**
      - **Found in *Bifidobacterium* and *Leuconostoc***

Glycolysis: The Embden-Meyerhoff-Parnas pathway



Glycolysis: The Hexose Monophosphate Pathway

Glucose

↓

Phosphogluconate

↓ Intercoversion of intermediates

5-C, 4-C, 7-C, 6-C, & 3-C

↓ ↓ Go to the EMP pathway

Pyruvic Acid + NADH + ATP

Glycolysis: The Entner-Doudoroff Pathway

Glucose

↓

Phosphogluconate

↓

Glyceraldehyde 3-P + Pyruvic Acid

Go to the EMP pathway

↓

Pyruvic Acid + NADH + ATP

Glycolysis: The Phosphoketolase Pathway

Glucose

↓

Phosphogluconate

↓

5-C intermediate + CO_2

↓

Glyceraldehyde 3-P + Acetyl CoA

↓ Go to the EMP pathway

Pyruvic Acid + NADH + ATP

C. Fermentation

1. Features of fermentation pathways
   a) Pyruvic acid is reduced to form reduced organic acids or alcohols.
   b) The final electron acceptor is a reduced derivative of pyruvic acid
   c) NADH is oxidized to form NAD: Essential for continued operation of the glycolytic pathways.
   d) O_2 is not required.
   e) No additional ATP are made.
   f) Gasses (CO_2 and/or H_2) may be released

C. Fermentation

2. **Fermentation pathways are useful as tools in biochemical identification.**
3. **Also used in industry: Synthesis of certain organic compounds.**

C. Fermentation

4. Examples of fermentation pathways

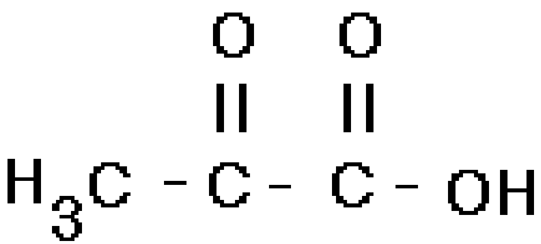
a) **Lactic acid fermentation**
   - Found in many bacteria;
     e.g. *Streptococcus sp, Lactobacillus acidophilus*

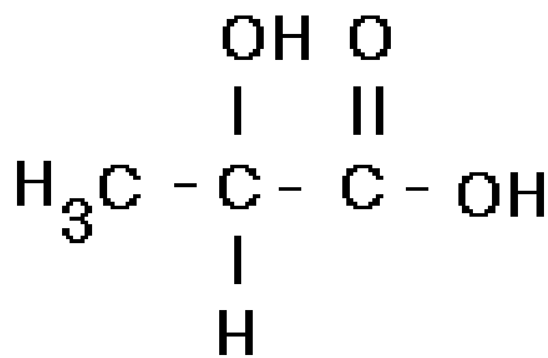
a) **Mixed acid fermentation**
   - e.g. *Escherichia coli*
   - Basis of the methyl red test

a) **2,3-Butanediol fermentation**
   - e.g. *Enterobacter aerogenes*
   - Basis of the Voges-Proskauer reaction

Fermentation Pathways: Lactic acid fermentation

Pyruvic Acid + NADH $\longrightarrow$ Lactic Acid + NAD





Fermentation Pathways: Mixed acid fermentation

Pyruvic Acid + NADH → Lactic Acid + NAD

→ Acetic Acid + Formic Acid

→ $H_2 + CO_2$

→ Carbonic Acid

Fermentation Pathways: Butanediol fermentation

2 Pyruvic Acid → Acetolactic acid + CO_2

→ Acetylmethylcarbinol + CO_2
(Acetoin)

+ NADH → 2,3-Butanediol + NAD

C. Fermentation

d) Other important fermentation end products

- **Ethanol**
  Saccharomyces cerevesiae
- **Propionic acid**
  Propionibacterium
- **Acetone, buteraldehyde, and butanol**
  Clostridium acetobutylicum

D. Respiration

1. **Features of respiratory pathways**
   a) **Pyruvic acid is oxidized completely to CO_2.**
   b) **The final electron acceptor is usually an inorganic substance.**
   c) **NADH is oxidized to form NAD: Essential for continued operation of the glycolytic pathways.**
   d) **O_2 may or may not be required.**
      - **Aerobic respiration: O_2 is the final e^- acceptor.**
      - **Anaerobic respiration: An substance, usually inorganic, other than O_2 is the acceptor (eg nitrate, nitrite, sulfate)**
   a) **A lot of additional ATP are made (up to 36 per glucose molecule)**

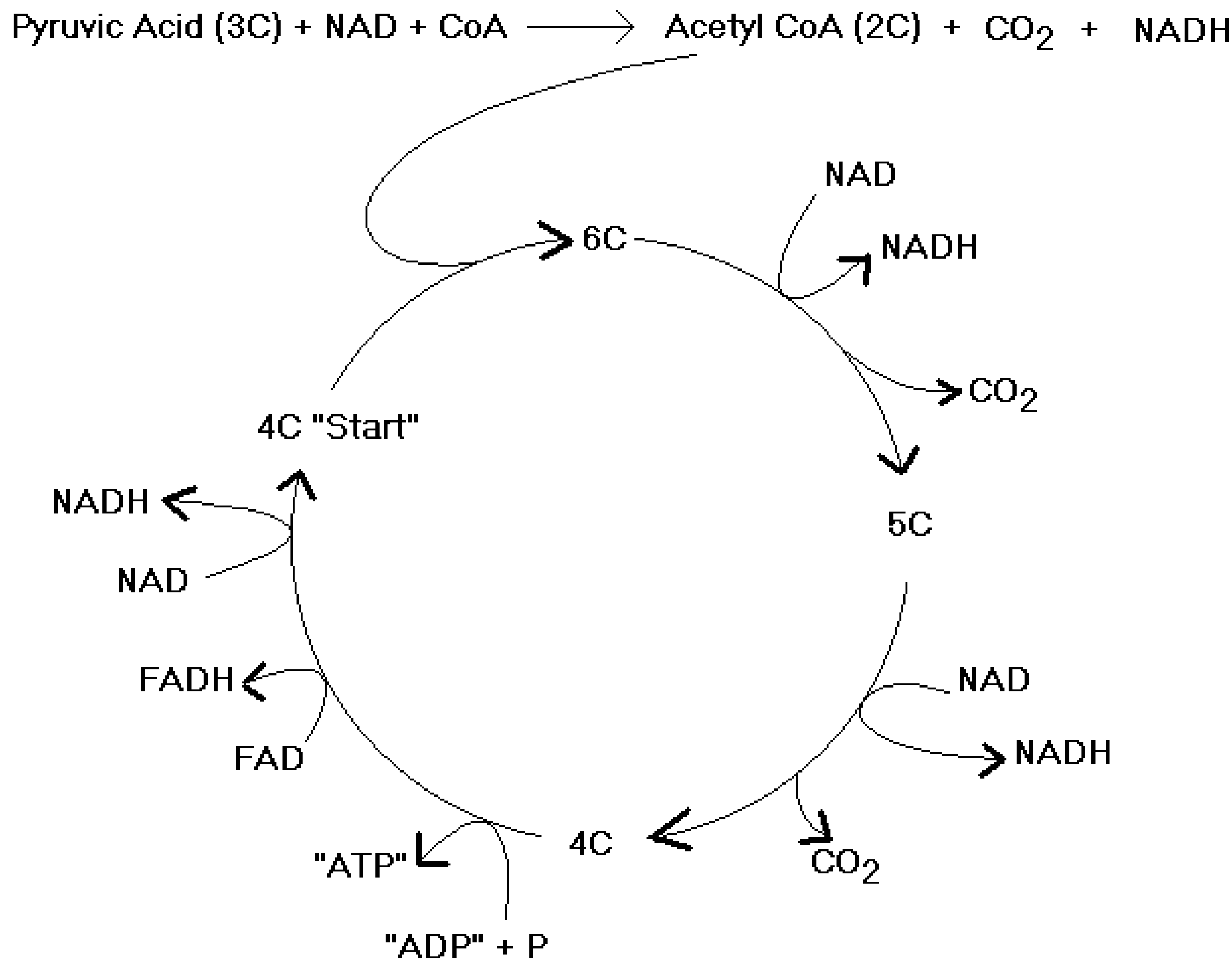
D. Respiration

2. Stages of Respiration

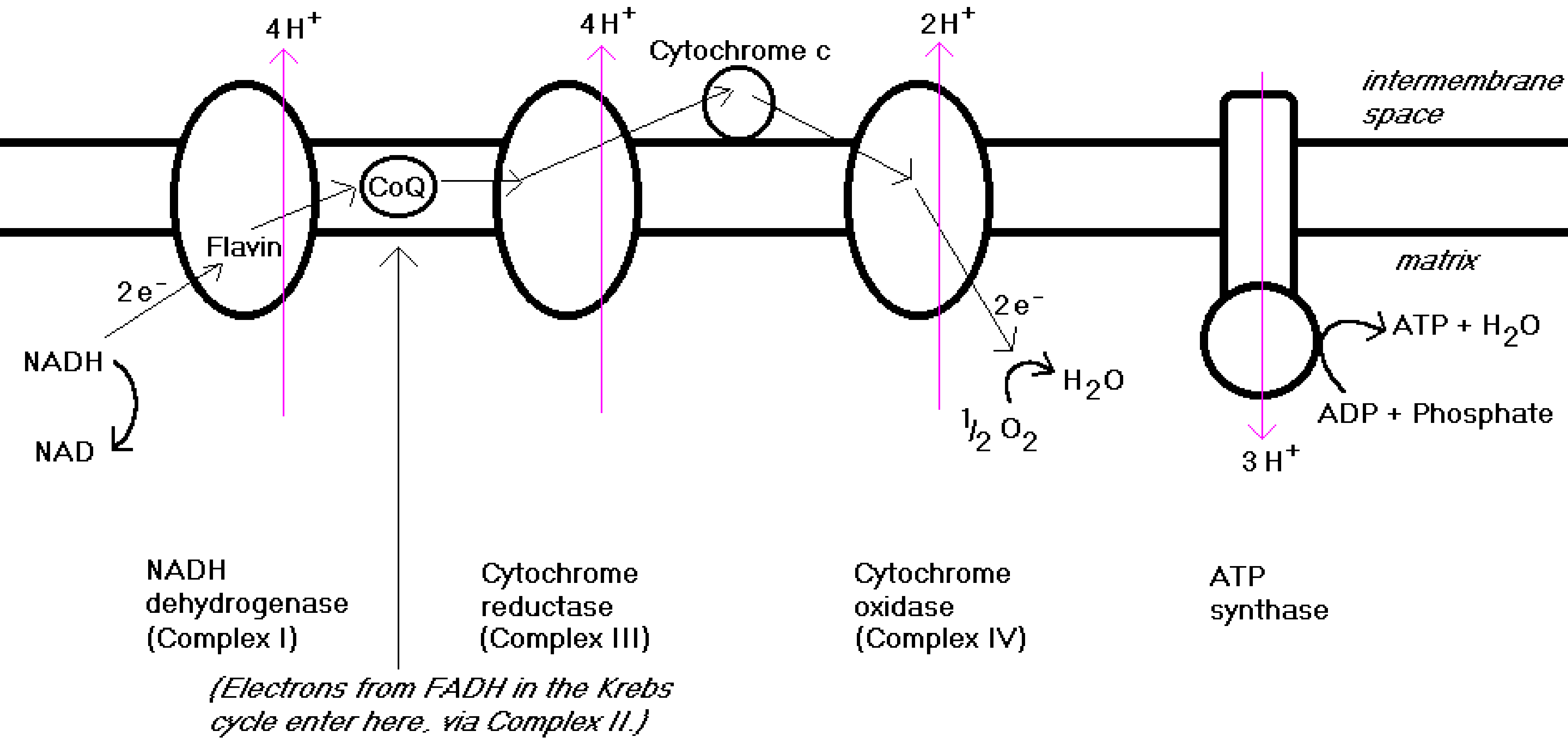
a) Preliminary reactions and the Krebs cycle (TCA or Citric Acid Cycle)

b) Respiratory electron transport

Respiration: Preliminary Reactions and the Krebs Cycle



Respiration: Electron transport in the mitochondria



Respiration: Different electron acceptors in bacteria

Aerobic respiration
(many bacteria & eukaryotic mitochondria)

$$\frac{1}{2}\,O_2 \rightarrow H_2O$$

Anaerobic respiration

$$NO_3^{2-} \rightarrow NO_2^{-}$$

Nitrate reduction
(e.g., in several gram-negative enteric bacteria such as E. coli)

$$NO_2^{-} \rightarrow N_2$$

Nitrite reduction
(e.g., Pseudomonas, Bacillus, and related soil and aquatic genera)

$$SO_4^{2-} \rightarrow H_2S$$

Sulfate reduction
(e.g., Desulfovibrio & Desulfotomaculatum)

$$S_o \rightarrow H_2S$$

Sulfur reduction
(Desulfuromonas)

E. Photosynthesis

1. Overview of Photosynthesis

a) Light-dependent Reactions:

- Light energy is harvested by photosynthetic pigments and transferred to special reaction center (photosystem) chlorophyll molecules.
- The light energy is used to strip electrons from an electron donor (the electron donor goes from a reduced to an oxidized state).
- The electrons are shuttled through a series of electron carriers from high energy state to a low energy state.
- During this process, ATP is formed.
- In the cyclic pathway of electron transport, electrons are returned to the electron transport chain
- In the noncyclic pathway, the electrons are used to reduce NAD (or NADP) to NADH (or NADPH)

E. Photosynthesis

b) Light-independent Reactions:

- **ATP and NADH (NADPH) from the light-dependent reactions are used to reduce CO_2 to form organic carbon compounds (carbon fixation).**
- **The reduced organic carbon is usually converted into glucose or other carbohydrates.**

E. Photosynthesis

2. Oxygenic photosynthesis

a) Found in cyanobacteria (blue-green algae) and eukaryotic chloroplasts

b) Electron donor is H_2O: Oxidized to form O_2

c) Two photosystems: PSII and PSI

d) Major function is to produce NADPH and ATP for the carbon fixation pathways

E. Photosynthesis

3. Anoxygenic photosynthesis

a) Found in:

- Green sulfur bacteria (e.g. *Chlorobium*)
- Green nonsulfur bacteria (e.g. *Chloroflexus*)
- Purple sulfur bacteria (e.g. *Chromatium*)
- Purple nonsulfur bacteria (e.g. *Rhodobacter*)

E. Photosynthesis

3. Anoxygenic photosynthesis (cont.)
   b) Electron donors vary:
      - H_2S or S_0 in the green and purple sulfur bacteria
      - H_2 or organic compounds in the green and purple nonsulfur bacteria
   c) Only one photosystem
      - In green bacteria, the photosystem is similar to PSI
      - In purple bacteria, the photosystem is similar to PSII
   d) Primary function is ATP production, chiefly via cyclic photophosphorylation

F. Chemolithotrophy

1. **Features of Chemolithotrophy**
   a) **Electrons are removed from a reduced inorganic electron donor**
   b) **The electrons are passed through a membrane-bound electron transport pathway, often coupled to the synthesis of ATP and NADH**
   c) **The electrons are ultimately passed to a final electron acceptor**
   d) **ATP and NADH may be used to convert CO_2 to carbohydrate**

F. Chemolithotrophy

2. **Examples of electron donors**
   a) Ammonia (NH_4^+) → Nitrite (NO_2^-)
      in *Nitrosomonas*
   b) Nitrite (NO_2^-) → Nitrate (NO_3^{2-})
      in *Nitrobacter*
   c) Hydrogen sulfide (H_2S) → Sulfur (S_0)
      in *Thiobacillus* and *Beggiatoa*
   d) Sulfur (S_0) → Sulfate (SO_4^{2-})
      in *Thiobacillus*
   e) Hydrogen (H_2) → Water (H_2O)
      in *Alcaligenes*

F. Chemolithotrophy

3. Examples of electron acceptors

a) Oxygen (O_2) $\rightarrow$ Water (H_2O) in many organisms

b) Carbon dioxide (CO_2) $\rightarrow$ Methane (CH_4) in the methanogenic bacteria

G. The Nitrogen Cycle

1. **Mineralization:**

   Organic nitrogen (mostly amino acids) $\rightarrow NH_4^+$

   (All organisms)

1. **Nitrification:**

   $NH_4^+ \rightarrow NO_2^-$ (*Nitrosomonas*)

   $NO_2^- \rightarrow NO_3^{2-}$ (*Nitrobacter*)

1. **Denitrification**

   $NO_3^- \rightarrow N_2O$

   $N_2O \rightarrow N_2$

   (Several species, including certain *Pseudomonas* and *Bacillus*)

G. The Nitrogen Cycle

4. Assimilatory Nitrate Reduction

$NO_3^{2-} \rightarrow$ Organic Nitrogen

(Many microbial species and plants)

4. N_2 fixation

$N_2 \rightarrow NH_4^+$

Free-living nitrogen fixers

eg *Azotobacter* and *Azospirillum*

Symbiotic nitrogen fizers

eg *Rhizobium* and *Bradyrhizobium*

Cyanobacteria attached to the cordgrass plant *Spartina* in salt marshes

Thank you