METABOLISM IN PROCARYOTES

Energy-Generating Metabolism

The term **metabolism** refers to the sum of the biochemical reactions required for energy generation and use of this energy to synthesize cell material from small molecules in the environment. Hence, metabolism which has an **energy-generating**, **through breakdown of complex component called catabolism**, and which has **an energy-consuming**, **through synthesis of complex component from simple ones called anabolism**. Catabolic reactions produce energy as **ATP**, which can be utilized in anabolic reactions to build cell material from nutrients in the environment. The relationship between catabolism and anabolism is illustrated in Figure below.



The relationship between catabolism and anabolism in a cell.

Importance of metabolism

- 1- Industrial; yogurt, cheese, bread.
- 2- Medical; diagnosis

3- Environment; cycling of elements, pollutant transformation, biodegradation.

ATP

During catabolism, useful energy is temporarily conserved in the "high energy bond" of **ATP - adenosine triphosphate**. No matter what form of energy a cell uses as its primary source, the energy is ultimately transformed and conserved as ATP. When energy is required during anabolism, it may be spent as ATP which has a value of about 8 kcal per mole. Hence, the conversion of ADP to ATP requires 8 kcal of energy, and the hydrolysis of ATP to ADP releases 8 kcal.



Mechanism of generating ATP

1-Substrate level phosphorylation (SLP) is the simplest, oldest and least-evolved way to make ATP. Here ATP is made during the conversion of an organic molecule from one form to another by transfer of phosphate group from a high- energy substrate to ADP. SLP occurs during fermentations and respiration (the TCA cycle), and even during some lithotrophic transformations of inorganic substrates.

2-Electron Transport Phosphorylation (ETP) or Oxidative

Phosphorylation is more complicated. It takes place during respiration, photosynthesis, lithotrophy and other types of bacterial metabolism. ETP requires that electrons removed from substrates by the electron carriers(NAD, FAD..) be dumped into an electron transport system (ETS) contained within a membrane, transferred to some final electron acceptor in the membrane (like O_2 in aerobic respiration), while their traverse through the ETS results in the extrusion of protons and the establishment of a **proton motive force (pmf)** across the membrane. An essential

component of the membrane for synthesis of ATP is a **membrane-bound ATPase**

NAD

Another coenzyme commonly involved in energy-producing metabolism, derived from the vitamin niacin, is the pyridine nucleotide, **NAD** (**Nicotinamide Adenine Dinucleotide**). The basis for chemical transformations of energy usually involves oxidation/reduction reactions. Oxidation: is the removal of electrons, Reduction: is the gain of electrons, Redox: is both.



Coenzyme A

Coenzyme A is another coenzyme frequently involved in energygenerating metabolism of procaryotes. The oxidations of pyruvate and alpha ketoglutarate, involving Coenzyme A, NAD, a dehydrogenation reaction and a decarboxylation reaction, are two of the most important, and complex, reactions in metabolism.

Heterotrophic Types of Metabolism

Heterotrophy (i.e. chemoheterotrophy) is the use of an organic compound (sugars from polysaccharide, amino acids from proteins,...) as a source of carbon and energy. We animals are familiar with heterotrophic metabolism. Many **Bacteria** (but just a few **Archaea**) are heterotrophs, particularly those that live in associations with animals. Heterotrophic bacteria are the masters of decomposition and biodegradation in the environment. Heterotrophic metabolism is driven mainly by two metabolic processes: fermentations and respirations.

Respiration

Respirations result in the **complete oxidation of the substrate** by an **outside electron acceptor**. It includes aerobic and anaerobic types.

Aerobic Respiration: consists basically of three stages:

1- glycolysis and/or other pathways 2- The tricarboxylic acid (TCA) cycle 3- A membrane and an associated electron transport system (ETS).

1-Glycolysis: The Embden-Meyerhof Pathway

The oxidation of glucose to pyruvic acid, produces ATP and NADH. Some end products of Embden-Meyerhof fermentations are essential components of foods and beverages, and some are useful fuels and industrial solvents. Diagnostic microbiologists use bacterial fermentation profiles in order to identify them, down to the genus level.



The Embden Meyerhof pathway for glucose dissimilation. The overall reaction is the oxidation of glucose to 2 pyruvic acid.

The first three steps of the pathway prime (phosphorylate) and rearrange the hexose for cleavage into 2 trioses (glyceraldehydephosphate). **Fructose 1,6-diphosphate aldolase** is the key (cleavage) enzyme in the E-M pathway. Each triose molecule is oxidized and phosphorylated followed by two substrate level phosphorylations that yield 4 ATP during the pathway to pyruvate.

Phosphoketolase Pathway(**Pentose phosphate pathway**)

This pathway is distinguished by the key cleavage enzyme, **phosphoketolase**, which cleaves pentose phosphate into glyceraldehyde-3-phosphate and acetyl phosphate. It is used by many bacteria include some species of *Lactobacillus* and *Leuconostoc*.



The heterolactic (phosphoketolase) pathway . Compare with the Embden-Meyerhof pathway . This pathway differs in the early steps before the cleavage of the molecule. The overall reaction in the fermentation of glucose is Glucose ------> Lactic acid + ethanol + CO_2 + 1 ATP (net).

The Entner-Doudoroff Pathway

Only a few bacteria, most notably *Zymomonas*, employ the Entner-Doudoroff pathway as a strictly fermentative way of life. However, many bacteria, especially those grouped around the pseudomonads, use the pathway as a way to degrade carbohydrates for respiratory metabolism. The E-D pathway yields 2 pyruvic acid from glucose (same as the E-M pathway) but like the phosphoketolase pathway, oxidation occurs before the cleavage, and the net energy yield is one mole of ATP per mole of glucose utilized.

2-Tricarboxylic acid (TCA) cycle

The **tricarboxylic acid (TCA) cycle** (also known as the **citric acid cycle** or the **Kreb's cycle**): It is used for the complete oxidation of the substrate. The end product is CO_2+H_2O

The pyruvate that is moved into the TCA cycle, eventually becoming oxidized to 3 CO_2 . Since 2 pyruvate are formed from one glucose, the cycle must turn twice for every molecule of glucose oxidized to 6 CO_2 .

Initially, pyruvate is oxidized and decarboxylated in a complex reaction involving NAD, Coenzyme A, and pyruvate dehydrogenase (pyruvate decarboxylase), forming the most central molecule in metabolism, Acetyl CoA. Acetyl CoA condenses with the 4C-compound, oxalacetic acid, intermediate, 6C-citric acid (citrate), a tricarboxylic acid. Citrate is isomerized to isocitrate, which is oxidized and decarboxylated forming alpha-ketoglutarate (akg). Alpha ketoglutarate dehydrogenase uses CoA and NAD to oxidize akg to succinyl CoA. Succinyl CoA is converted to succinate during a substrate level phosphorylation yielding GTP (equivalent to ATP). This completes the decarboxylation of pyruvate forming 3 CO_2 . The remaining three steps in the cycle complete the oxidation of succinate and regenerate the oxalacetate. During the oxidation of pyruvic acid to 3 CO₂ by one turn of the TCA cycle, 4 NADH2, 1 FADH2 and one ATP (actually GTP) are produced. Since the TCA cycle is an important amphibolic pathway, several intermediates of the cycle may be withdrawn for anabolic (biosynthetic) pathways.



3-A membrane and an associated electron transport system (ETS).

The ETS is a **sequence of electron carriers in the plasma membrane** that transports electrons taken from the substrate through the chain of carriers to a final electron acceptor. This releases energy that can be harvested by the cells in the process of ATP synthesis by the mechanisms of **electron transport phosphorylation**. The operation of the ETS establishes a proton motive force (pmf) due to the formation of a proton gradient across the membrane.

An **outside electron acceptor**. For **aerobic respiration** the electron acceptor is O_2 , of course. Molecular oxygen is reduced to H_20 in the last step of the electron transport system. But in the bacterial processes of **anaerobic respiration**, the final electron acceptors may be SO_4 or S or NO_3 or NO_2 or certain other inorganic compounds.

The overall reaction for the aerobic respiration of glucose is

Glucose + 6 O₂ -----> 6 CO₂ + 6 H₂0 + 38 ATP + 688 kcal (total)

The diagram below of aerobic respiration integrates these metabolic processes into a scheme that represents the overall process of respiratory metabolism. A substrate such as glucose is completely oxidized to to CO_2 by the combined pathways of glycolysis and the TCA cycle. Electrons removed from the glucose by NAD are fed into the ETS in the membrane. As the electrons traverse the ETS, a pmf becomes established across the membrane. The electrons eventually reduce an outside electron acceptor, O_2 , and reduce it to H_2O . The pmf on the membrane is used by the ATPase enzyme to synthesize ATP by a process referred to as "oxidative phosphorylation".



Model of aerobic respiration. Glucose is oxidized to CO_2 via the TCA cycle. Most electrons are removed from the glucose by NAD and donated to the electron transport system in the cell membrane. The ultimte electron acceptor is O_2 which becomes reduced to H_2O . As a result of the electron transport process, pmf is established on the membrane . pmf drives the synthesis of ATP during the process of oxidative phosphorylation.

Fermentation

Fermentation is metabolism in which energy is derived from the **partial oxidation of an organic compound** using **organic intermediates as electron donors and electron acceptors**. **all ATP is produced by substrate level phosphorylation**.



Lactic acid bacteria reduce the pyruvate to lactic acid (lactate); yeast reduce the pyruvate to alcohol (ethanol) and $\rm CO_2$.

Besides lactic acid, Embden-Meyerhof, Phosphoketolase and Entner-Doudoroff Pathway, fermentations in bacteria can lead to a wide array of end products depending on the pathways taken in the reductive steps after the formation of pyruvic acid. Some of other pathways proceeding from pyruvic acid in certain bacteria. Usually, these bacterial fermentations are distinguished by their end products into the following groups.

1. **Homolactic Fermentation**. **Lactic acid** is the sole end product. Pathway of the homolactic acid bacteria (*Lactobacillus, Lactococcus* and most streptococci).

2. Mixed Acid Fermentations. Mainly the pathway of the *Enterobacteriaceae*. End products are a mixture of **lactic acid**, **acetic acid**, **formic acid**, **succinate** and **ethanol**, with the possibility of gas formation (CO_2 and H_2).

3. Butanediol Fermentation. 4. Butyric acid fermentations,

5. Butanol-acetone fermentation. 6. Propionic acid fermentation.