

# **CELLULAR RESPIRATION (THE ROLE OF MITOCHONDRIA IN CELLULAR RESPIRATION)**

## **I. INTRODUCTION**

Mitochondria are organelles present in all aerobic cells. Their essential role is to store the energy released by the enzymatic oxidation of nutrient molecules in the form of adenosine triphosphate (ATP).

Mitochondria utilize pre-synthesized organic molecules of diverse types (pyruvate, fatty acids, amino acids). Through complete oxidation using atmospheric oxygen, they extract all the energy contained within these chemical bonds; this process is known as cellular respiration.

The Krebs cycle, which takes place in the mitochondrial matrix, ensures the complete degradation of acetyl residues and provides CO<sub>2</sub> and reducing power in the form of reduced coenzymes: NADH + H<sup>+</sup> and FADH<sub>2</sub>. These coenzymes feed into a redox chain (electron transport chain). The stages of this chain, which occur primarily at the level of the inner mitochondrial membrane, are responsible for providing energy in the form of ATP.

In the following sections, we will detail these different modes of energy conversion and the physiological role of mitochondria in this operation within eukaryotic cells.

## **II. MITOCHONDRIA AND AEROBIC RESPIRATION**

### **II. 1. Composition of Mitochondrial Membranes**

The structural and molecular organization of mitochondria is identical across all eukaryotes, including both animals and plants. They generally take the form of rods with rounded ends, with a diameter of approximately 0.5 μm and a length ranging from one to several microns.

The outer membrane is a lipid bilayer highly enriched in a protein called porin. This protein forms pores that allow the passage of molecules with a molecular mass of up to 6 kDa; consequently, this membrane is highly permeable to all metabolites. However, it does not allow the passage of protons (H<sup>+</sup>), which accumulate in the intermembrane space.

The inner membrane is also a lipid bilayer and features numerous folds known as mitochondrial cristae. The number and surface area of these cristae are correlated with the cell's ATP demand. For example, in cardiac cell mitochondria (where ATP demand is very high), the number of cristae is three times higher than in those of hepatocytes.

The inner membrane exhibits low fluidity, and its protein and lipid composition is highly specialized, both: Quantitatively: Proteins (70% to 80%) and lipids (20% to 30%) and Qualitatively: It is devoid of cholesterol but contains a unique phospholipid, diphosphatidylglycerol (cardiolipin), a molecule that only this organelle is capable of synthesizing.

In addition to the protein complexes of the respiratory chain, the inner membrane features about ten different permeases that perform active co-transport between the intermembrane space and the matrix for: ATP, ADP, pyruvate, fatty acids, and phosphate ions.

## **II.2. Complete Degradation of Glucose in the Cell**

### **II.2.1. Glycolysis**

- The degradation of glucose begins in the cytoplasm via glycolysis, a stage that does not require the presence of O<sub>2</sub> ("anaerobic respiration").
- Glycolysis facilitates the formation of two pyruvate molecules from a single glucose molecule, with the production of 2 ATP and 2NADH + H<sup>+</sup>.

### **II.2.2. The Krebs Cycle**

- Pyruvate is transported into the mitochondrion and converted into Acetyl CoA through an oxidation reaction that reduces NAD<sup>+</sup> to NADH + H<sup>+</sup> and a decarboxylation reaction.
- Acetyl CoA enters the citric acid cycle, where it is decarboxylated and dehydrogenated, yielding FADH<sub>2</sub>, NADH + H<sup>+</sup>, ATP, and CO<sub>2</sub>.
  - 2 decarboxylation reactions (x2).
  - 3 oxidations that reduce NAD<sup>+</sup> to NADH + H<sup>+</sup> (x2).
  - 1 oxidation that reduces FAD to FADH<sub>2</sub> (x2).
  - Production of 1 ATP (x2).
- NADH and FADH<sub>2</sub> are activated intermediates that carry high-energy electrons; their oxidation occurs via the respiratory chain (electron transport chain).
- NAD<sup>+</sup> is nicotinamide adenine dinucleotide (a derivative of vitamin B3).

NAD<sup>+</sup> is the oxidized form. NADH is the reduced activated form. NADH is a highly reducing molecule.

- FAD is flavin adenine dinucleotide.

FAD is the oxidized form. FADH<sub>2</sub> is the reduced activated form. FADH<sub>2</sub> contains riboflavin, which is derived from vitamin B<sub>2</sub>.

### **II.3. Oxidative Phosphorylation**

This is the final stage of catabolism (the complete degradation of glucose), during which the majority of the metabolic energy required by the cell is produced (approximately 34 molecules of ATP).

The energy released by electrons (where NADH + H<sup>+</sup> and FADH<sub>2</sub> serve as the primary electron donors) is used for:

- The translocation of protons (H<sup>+</sup>) from the matrix to the intermembrane space through the complexes of the respiratory chain (Complexes I, III, and IV).
- The phosphorylation reactions of ADP into ATP (via ATP synthase). Inorganic phosphate, in the form of H<sub>2</sub>PO<sub>4</sub><sup>-</sup> from the environment, is incorporated into the ADP (the inner mitochondrial membrane is rich in H<sup>+</sup>/H<sub>2</sub>PO<sub>4</sub><sup>-</sup> symporters that import the Pi).

If electron transport is inhibited, ADP phosphorylation does not occur. This demonstrates the existence of energy coupling between respiratory oxidations (respiratory chain complexes) and the phosphorylation of ADP into ATP (ATP synthase).

## **III. THE RESPIRATORY CHAIN**

This is the electron transport system in which electron pairs are bound and transferred by specialized carriers (proteins within the inner mitochondrial membrane) until they combine with oxygen to form water.

It consists of a sequence of redox reactions that facilitate the transfer of high-energy electrons from reduced activated molecules (NADH and FADH<sub>2</sub>) to molecular oxygen, resulting in the production of ATP and water. At the end of the chain, these electrons are accepted by Oxygen (O<sub>2</sub>), which combines with protons (H<sup>+</sup>) from the environment to form water (2H<sup>+</sup> + 2 e<sup>-</sup> + O<sub>2</sub> = H<sub>2</sub>O).

The energy lost by the electrons and the presence of  $H^+$  protons activate ATP synthase, which is localized in the inner mitochondrial membrane. This enzyme catalyzes the production of ATP in large quantities.

*Note:* Each mitochondrial  $NADH + H^+$  molecule generates 3 ATP as it passes through the cytochrome chain, while each  $FADH_2$  molecule generates 2 ATP.

### III.1. GENERAL ORGANIZATION OF THE RESPIRATORY CHAIN

- There are four multienzyme complexes, numbered I through IV, which facilitate electron transport.
- Three of these complexes (I, III, and IV) function as proton pumps.
- These complexes are linked by two mobile carriers:
  - 1) Ubiquinone (also known as Coenzyme Q or UQ): This is a lipid molecule.
  - 2) Cytochrome c: This is a heme protein. It is soluble within the intermembrane space and transports a single electron at a time.
- There is a fifth complex: ATP synthase.

#### III.1. 1. Complex I: NADH-CoQ Reductase

##### (NADH Dehydrogenase / NADH-Ubiquinone Oxidoreductase)

This is the largest transmembrane complex in the respiratory chain. It is a flavoprotein associated with a non-protein prosthetic group, **Fe-S** (where sulfur stabilizes the iron atoms that transfer electrons). This complex catalyzes the transfer of electrons from the donor,  **$NADH + H^+$** , to the acceptor, **CoQ (Ubiquinone)**.

Two electrons are first transferred to **FMN** (Flavin Mononucleotide), converting it into **FMNH<sub>2</sub>**. The transfer then continues to the **Fe-S** group, which picks up one electron at a time, transitioning from the ferric state ( $Fe^{+++}$ ), oxidized) to the ferrous state ( $Fe^{++}$ , reduced). Finally, the electrons are transferred to **CoQ**, which is lipid-soluble, hydrophobic, and mobile within the mitochondrial membrane. The energy released during this process is sufficient to pump **4H<sup>+</sup>** from the matrix into the intermembrane space.

### **III.1.2. Complex II: Succinate-CoQ Reductase**

#### **(Succinate-Ubiquinone Oxidoreductase)**

This is the smallest protein complex in the respiratory chain. It is located on the matrix side of the membrane and contains few proteins. It catalyzes the conversion of succinate to fumarate within the Krebs cycle. Two electrons are transferred via FAD to the Fe-S protein (  $\text{Fe}^{+++}$  is reduced to  $\text{Fe}^{++}$ ), and the transfer continues to CoQ. The energy released here is not sufficient to transport protons; therefore, this complex does not directly contribute to the proton gradient used for ATP synthesis.

### **III.1.3. Complex III: CoQ-Cytochrome c Reductase**

This complex contains two types of cytochrome b ( b562 and b566, named after their absorption wavelengths). Electrons pass from cytochrome b566 to b562, then through an Fe-S center to cytochrome c1, and finally to cytochrome c.

Complex III catalyzes the electron transfer between CoQ (  $\text{QH}_2$ , reduced) and cytochrome c, both of which are mobile carriers. It oxidizes  $\text{UQH}_2$  into  $\text{UQ}$  and reduces cytochrome c. Notably, cytochrome c accepts only one electron at a time. The energy from this electron transfer is sufficient for the translocation of  $4\text{H}^+$  into the intermembrane space.

### **III.1.4. Complex IV: Cytochrome c Oxidase**

This complex retrieves electrons from Complex III. It consists of two types of hemes:

- Heme a, which transfers electrons to the copper center  $\text{Cu}_A$ .
- Heme  $a_3$ , which receives electrons from  $\text{Cu}_A$  and transmits them to the copper center  $\text{Cu}_B$ .

The energy released is sufficient to transfer  $2\text{H}^+$  from the matrix to the intermembrane space. This complex catalyzes the final reduction of molecular oxygen into water:

### **III.1.5. Complex V: ATP Synthase (ATPosome)**

Since the inner membrane is impermeable to ions, the only way for the protons accumulated in the intermembrane space to re-enter the matrix is through specialized transmembrane channels called ATP synthases. This complex consists of:

- The F1 particle: A spherical head that functions like a turbine activated by the flow of protons, enabling the synthesis of ATP. F1 is located within the matrix.

- The F<sub>0</sub> particle: A transmembrane stalk or channel that acts as the proton pore, anchoring the complex in the membrane.

The ATP synthase complex facilitates the return of protons to the matrix, which ultimately powers the synthesis of ATP.

### III.2. Respiratory Chain Inhibitors

**Table : Names and Mechanisms of Action of Respiratory Chain Inhibitors**

Inhibition Type	Action	Examples
<b>Oxidation Inhibitors</b>	Blockage of electron transfer	<b>Cytochrome oxidase inhibitors:</b> - <b>CO</b> (binds to Fe <sup>2+</sup> ) - <b>Cyanide</b> (binds to ferric iron Fe <sup>3+</sup> )
<b>Phosphorylation Inhibitors (Uncouplers)</b>	No ATP synthesis; energy released as heat	- <b>2,4-dinitrophenol (DNP):</b> A poison - <b>Thermogenin:</b> A physiological compound

2,4-dinitrophenol (DNP) diffuses across the inner mitochondrial membrane and acts as a proton carrier. By doing so, it collapses the proton gradient associated with electron transport. Uncouplers inhibit phosphorylation without disrupting electron transport. In this case, the free energy provided by electron transport is entirely dissipated as heat. This heat production in the presence of an uncoupler is known as thermogenesis.

Animals and humans possess a specialized adipose tissue called brown fat. This tissue is highly enriched in mitochondria that contain a specific uncoupling protein in their inner membrane called thermogenin. Once the gradient is established, the protons—instead of returning to the matrix through the F<sub>0</sub> channel—are channeled through thermogenin to produce heat rather than ATP. This process is essential for thermoregulation in cold environments and for maintaining body temperature in hibernating animals.