## **Respiration in Plants - Part 2**

## Objectives

After going through this lesson, the learners will be able to understand the following:

- Aerobic Respiration
- Oxidative Decarboxylation of Pyruvate The Link Between Glycolysis and Krebs Cycle
- Steps of the Krebs Cycle
- The Electron Transfer Chain
- Oxidative Phosphorylation

## **Content Outline**

- Aerobic Respiration
- Oxidative Decarboxylation Of Pyruvate
- Krebs Cycle
- The Electron Transfer Chain
- Oxidative Phosphorylation

### **Aerobic Respiration**

Aerobic respiration is the process that leads to a complete oxidation of organic substances in the presence of oxygen, and releases carbon-dioxide, water and a large amount of energy present in the substrate. Their action may be represented as follows when glucose is the substrate.

# C6H12O6 + 6O2 Enzymes 6CO2 + 6H2O + 686 kcal/2870 kJ

This type of respiration is most common in higher organisms.

The organisms which carry on this type of respiration are called aerobes.

Aerobic respiration is carried out in 2 steps-

 Oxidative decarboxylation of the pyruvate formed in glycolysis to produce acetyl Coenzyme A and subsequent stepwise oxidation of the acetyl CoA by the Krebs cycle to produce carbon-dioxide, NADH FADH and ATP. These reactions take place in the mitochondrial matrix. 2. The passage of the electrons through a series of electron acceptors forming the electron transfer chain (located in the inner membrane of the mitochondria) to molecular oxygen resulting in regeneration of NAD and FAD with simultaneous synthesis of ATP which is formed by oxidative phosphorylation.

### **Oxidative Decarboxylation of Pyruvate**

When oxygen is present i.e. under aerobic conditions, pyruvate produced in the cytosol as the end product of glycolysis, is transported into the mitochondrial matrix.

In the mitochondrial matrix the pyruvate undergoes oxidative decarboxylation by a complex set of reactions catalysed by pyruvate dehydrogenase. The reactions catalysed by pyruvate dehydrogenase require the participation of several factors, including Mg ions, thiamine pyrophosphate (TPP), lipoic acid, NAD+ and Coenzyme A. The reaction is as follows (figure 1)-

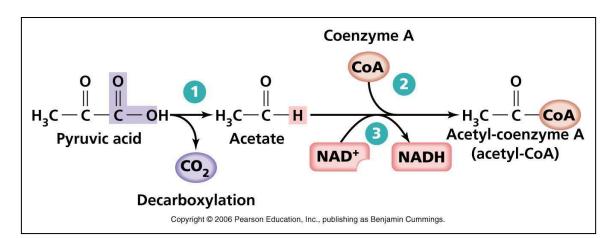


Figure 1: Oxidative decarboxylation of pyruvic acid

Carbon dioxide, NADH and acetyl Coenzyme A are produced as follows -

- 1. A carboxyl group is removed from pyruvate and released as carbon dioxide.
- 2. The two-carbon molecule from the first step is oxidized, and NAD<sup>+</sup> accepts the electrons to form NADH.
- 3. The oxidized two-carbon molecule, an acetyl group, is attached to Coenzyme A to form acetyl CoA.

The acetyl CoA then enters a cyclic pathway called the tricarboxylic acid cycle (TCA cycle) which is also known as the citric acid cycle (as the first product of the cycle is citric acid) or Krebs' cycle after the scientist Hans Krebs who first elucidated it.

Acetyl CoA is the link between glycolysis and the TCA cycle.

## Krebs Cycle / TCA Cycle / Citric Acid Cycle

The citric acid cycle (Figure 2) starts with the condensation of acetyl group with oxaloacetic acid (OAA) and water to yield citric acid (Figure 2). The reaction is catalysed by the enzyme citrate synthase and a molecule of CoA is released.

Citrate is then isomerised to isocitrate. It is followed by two successive steps of decarboxylation, leading to the formation of - ketoglutaric acid and then succinyl - CoA.

In the remaining steps of the citric acid cycle, succinyl-CoA is oxidised to OAA allowing the cycle to continue. During the conversion of succinyl-CoA to succinic acid a molecule of GTP is synthesised. **This is a substrate level phosphorylation**. In a coupled reaction GTP is converted to GDP with the simultaneous synthesis of ATP from ADP.

There are three points in the cycle where  $NAD^+$  is reduced to  $NADH^+ H^+$  and one point where  $FAD^+$  is reduced to  $FADH_2$ .

The continued oxidation of acetyl CoA via the TCA cycle requires the continued replenishment of oxaloacetic acid, the first member of the cycle.

In addition it also requires regeneration of NAD<sup>+</sup> and FAD<sup>+</sup> from NADH and FADH<sub>2</sub> respectively.

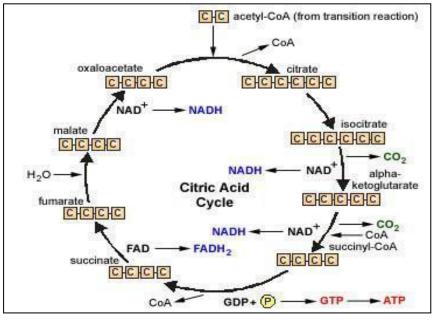


Figure 2: Krebs cycle/TCA cycle/Citric acid cycle

The summary equation for the complete oxidation of one molecule of pyruvic acid in the TCA cycle is as follows

Pyruvic acid+  $4NAD+FAD+H_2O + ADP+Pi= 3CO_2 + 4NADH + 4H + FADH_2 + ATP$ Since, one molecule of glucose was broken down in glycolysis to produce two molecules of pyruvic acid, the overall product from the breakdown of the I molecule of glucose is 6 molecules of carbon dioxide, 8 molecules of NADH, 2 molecules of FADH\_2 and 2 GTP.

### **Electron Transfer Chain**

The NADH and FADH<sub>2</sub> produced in the Krebs cycle enter the electron transport chain. The electron transport chain catalyses a flow of electrons from NADH and FADH<sub>2</sub> to oxygen. The role of the electron transport chain is to oxidise NADH and FADH<sub>2</sub> and thereby regenerate NAD and FAD to replenish the cellular pool of these reductants so that the Krebs cycle remains functional. The free energy released in the oxidation of NADH and FADH<sub>2</sub> is used to generate an electrochemical proton gradient across the inner membrane of the mitochondria which as we shall see is essential for ATP synthesis.

The electron transport system has electron transport proteins organised to form four complexes (complexes I to IV) and two mobile electron carriers ubiquinone and cytochrome c (Figure 3). Complex V is involved in ATP synthesis (Figure 3).

<u>Complex I</u>: (NADH dehydrogenase). NADH generated in the mitochondrial matrix in the TCA cycle is oxidised by Complex I. Complex I transfers electrons from NADH to ubiquinone. Ubiquinone is not associated with any protein and can move within the membrane. The reduced ubiquinone delivers the electrons picked up from Complex I to Complex III and gets oxidised in the process.

<u>Complex II</u>: (Succinate dehydrogenase). FADH<sub>2</sub> produced during conversion of succinate to fumarate in the TCA cycle, is oxidised by this complex. Complex II transfers electrons from FADH<sub>2</sub> to ubiquinone and the reduced ubiquinone in turn transfers the electrons to Complex III.

<u>Complex III</u>: (cytochrome  $bc_1$  complex or Cytochrome c reductase). This complex oxidises reduced ubiquinone and transfers the electrons via the cytochrome  $bc_1$  complex to cytochrome c. Cytochrome c is mobile within the membrane and transports electrons from Complex III to Complex IV.

<u>Complex IV</u>: (Cytochrome c oxidase). Complex IV is the terminal oxidase and brings about the four electron reduction of a molecule of oxygen to two molecules of water. Oxygen is the ultimate electron acceptor in the electron transfer chain.

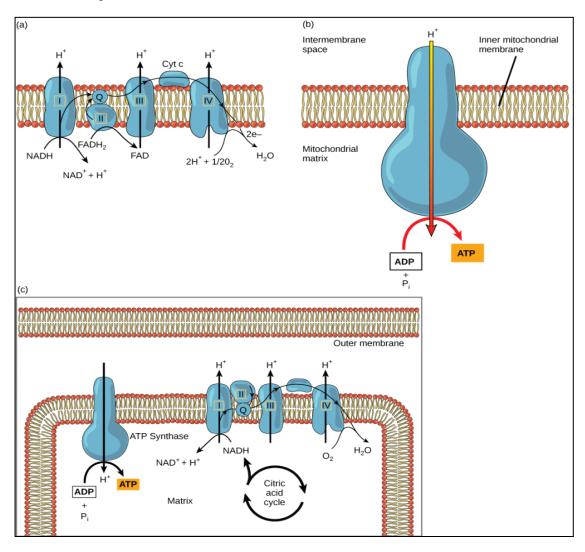


Figure 3

**3a.** The four multiprotein complexes and two mobile carriers( Q= ubiquinone and cyt c= cytochrome c) in the inner membrane of the mitochondria .

**3b.** Complex V Involved in ATP synthesis located in the inner membrane of the mitochondria.

**3c.** Organisation of the electron transport system in the inner membrane of mitochondria and ATP synthesis (oxidative phosphorylation) in the mitochondrial matrix.

<u>Complex V</u>: This complex consists of two major components, a transmembrane proton channel designated as F0 and an enzymatic domain within the mitochondrial matrix

designated as F1. The enzyme ATP synthase is associated with  $F_1$  and is involved in the synthesis of ATP from ADP and Pi.

Thus the passage of electrons in the electron transport chain is coupled to ATP synthesis in Complex V.

#### **Oxidative Phosphorylation**

Mitochondrial ATP synthesis is called oxidative phosphorylation as it is associated with oxygen consumption (we have already seen that oxygen is the terminal electron acceptor in the electron transfer chain).

In the course of mitochondrial electron transport, protons are extruded from the mitochondrial matrix into the inter membrane space (space between the inner and outer membranes of the mitochondria – Figure 3c). Proton extrusion takes place at three locations associated with Complex I, III and IV. The accumulation of protons in the intermembrane space results in a proton disequilibrium across the inner membrane. This generates a proton motive force across the membrane. ATP synthesis is driven by the return of protons to the mitochondrial matrix through the integral membrane protein complex called ATP synthase/ coupling factor /  $F_0$ - $F_1$  – ATPase (referred to as complex V in the electron transport system).

Unlike photophosphorylation where light energy is used for the production of the proton gradient required for phosphorylation, in respiration it is the energy of oxidation-reduction that is utilised for ATP production. It is for this reason that the process is called oxidative phosphorylation.

It was Peter Mitchell who first proposed that a proton motive force established across a membrane is responsible for ATP production in respiration. This is known as the chemiosmotic theory for ATP production.

In the next module we shall try to take stock of the number of ATP molecules produced by oxidation of a molecule of glucose.