4

Respiration

Every organism carries out various life processes. All these processes are collectively called physiological processes. A constant supply of energy is essential to carry out these processes. Living organisms derive this energy by oxidation of macromolecules that we call food. Only green plants and cyanobacteria can synthesis their own food by the process of photosynthesis. Even in green plants only those organs which possess chloroplast perform the photosynthesis but food is required for the oxidation by all the organs even, which do not possess chloroplasts. Hence, synthesized food is translocated to all non green parts. Most important part is that all the food that respired for life processes comes from photosynthesis. Thus, energy exchange takes place during biological processes. Based on the energy exchange there are two types of biological processes : (1) Endergonic processes and (2) exergonic processes.

Energy is stored during **endergonic process,** whereas energy is released during **exergonic process.** Photosynthesis is an endergonic process whereas respiration is an exergonic process. The breakdown of C-C bonds of complex compounds through oxidation within the cells releasing considerable amount of energy is called respiration. Since it takes place in cell, it is also known as cellular respiration. The substances which are oxidized during this process are known as respiratory substrates. Both plants and animals respire. Respiration is an important physiological process taking place in the mitochondria of the cells of all organisms. A part of the energy released during this process is utilized for the synthesis of ATP. The organic nutrients synthesised by the organisms or taken in as food undergo slow combustion during this process.

Do plants breathe ?

Plants require oxygen for respiration to occur and they also give out CO_2. However, plants, unlike animals, do not have specialized respiratory organs. However, stomata and lenticels are present which take part in gaseous exchange. There are several reasons why plants do not have specialized respiratory organs, such as –

- Each plant part is directly involved in the exchange of gases.
- As plant parts respire at rates far lower than the animals do, the need of gas exchange is not much.

- When cells photosynthesise, availability of O_2 is not a problem in these cells since O_2 is released within cell.

The strategy that the plant cell uses it to catabolise the glucose molecule in such a way that not all the liberated energy goes out as heat. During respiration, the stored energy in glucose is gradually released through controlled oxidation. The released energy is stored by forming ATP from ADP. The entire process of respiration can be represented by the following equation.

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

Glycolysis

The break down of glucose to pyruvic acid is called glycolysis. The term glycolysis has originated from the Greek words, 'glycos' for 'sugar' and 'lysis' for 'splitting'. The scheme of glycolysis was discoverd by Embden, Meyerhof and Parnas and is often known as EMP pathway.

Glycolysis is common to all living organisms. Both, aerobic as well as anaerobic respiration begin with glycolysis. In this phase O_2 is not utilized. Glycolysis occurs in the cytoplasm matrix of cells. During glycolysis glucose is converted into two (2) molecules of pyruvic acid. In plants, glucose is derived from sucrose which is synthesized during photosynthesis. The enzyme invertase converts sucrose into glucose and fructose, which readily enter the glycolytic pathway.

In this phase, first of all, the molecule of glucose is phosphorylated to Glucose-6 phosphate with the use of ATP. The phosphate liberated from ATP is joined to carbon number 6 of glucose. ATP becomes ADP. The enzyme, responsible is hexokinase.

Glucose-6-phosphate is converted into its isomer fructose-6-phosphate, through molecular rearrangement. Now, fructose-6 phosphate is again phosphorylated with the help of ATP forming Fructose-1-6-biphosphate. During this ATP is converted to ADP.

Fructose-1-6-diphosphate splits and two 3-C molecules – phosphoglyceraldehyde (PGAL) and dihydroxyacetone phosphate (DHAP) come into existance. Both these triose sugars are interconvertible. Normally, dihydroxyacetone phosphate is converted into phosphoglyceraladehyde. Thus two molecules of 3- phosphoglyceraldehyde are formed.

Energy will now be released in the subsequent phase. Till now, 2 molecules of ATP are consumed.

Each molecule of 3- phosphoglyceraladehyde is oxidized and $2H^+$ and $2e^-$ are released from it. The released components are accepted by NAD forming NADH + H^+. Simultaneously, a molecule of inorganic phosphate is also added. As this happen, 1,3 biphosphoglyceric acid (BPGA) is formed.

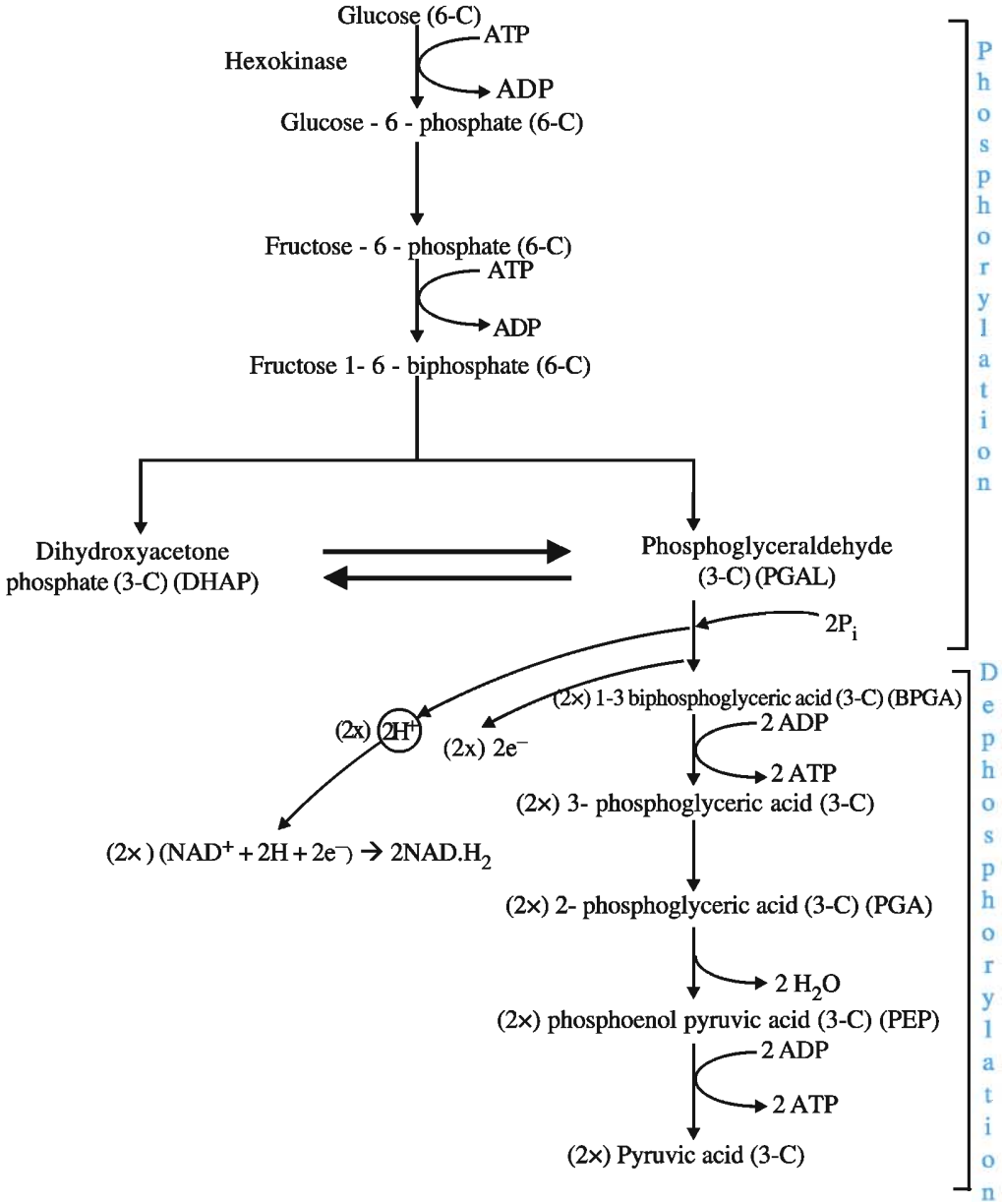
This phase of reactions constitutes phosphorylation phase. Now, the phase of dephosphorylation begins.

1,3- biphosphoglyceric acid is dephosphorylated and it is converted to 3-phosphoglyceric acid (PGA). The phosphate thus released from the substrate combines with ADP and forms ATP. This is called substrate phosphorylation.

Now, 3-phosphoglyceric acid is first converted into, 2-phosphoglyceric acid and then into phosphoenol pyruvic acid (PEP). H_2O is released during this process.

Phosphoenol pyruvic acid is dephosphorylated and converted into pyruvic acid. The phosphate released from the substrate combines with ADP and forms ATP.

Thus, at the end of entire process, two molecules of pyruvic acid are formed from one molecule of glucose. Two molecules of ATP are consumed and four molecules are formed. Thus, there is a net gain of 2 ATP. 2 molecules of $NADH_2$ are also formed. Now there are three major ways in which different cells utilize pyruvic acid produced during glycolysis. These are lactic acid fermentation, alcoholic fermentation and aerobic respiration.



Glycolysis

Fermentation

When respiration takes place in the absence of molecular oxygen, it is said to be anaerobic respiration. It results in incomplete breakdown of glucose. During this process CO_2 and organic compounds like ethyl alcohol and lactic acid are produced and some energy is released. Water molecule is not produced during anaerobic respiration. This reaction is also called fermentation. The fermentation are of two types :

Alcoholic fermentation : This fermentation occurs in Yeast. The incomplete oxidation of glucose is achieved under anaerobic conditions, where pyruvic acid is converted to CO_2 and ethanol. The enzymes pyruvic acid decarboxylase and alcohol dehydrogenase catalyse these reactions.

The pyruvic acid is first decarboxylated to acetaldehyde in the presence of enzyme pyruvic acid decarboxylase and thiamine pyro-phosphate (TPP).

$$2CH_3CO\ COOH \xrightarrow{\text{decarboxylase + TPP}} 2CH_3CHO + 2CO_2$$

Acetaldehyde

Acetaldehyde in the presence of the enzyme acetaldehyde alcohol dehydrogenase and coenzyme $NADH_2$ is reduced to ethyl alcohol. $NADH_2$ is oxidized to NAD.

$$2CH_3CHO + 2\ NADH_2 \xrightarrow{\text{alcohol dehydrogenase}} 2\ (C_2H_5OH) + 2\ NAD$$

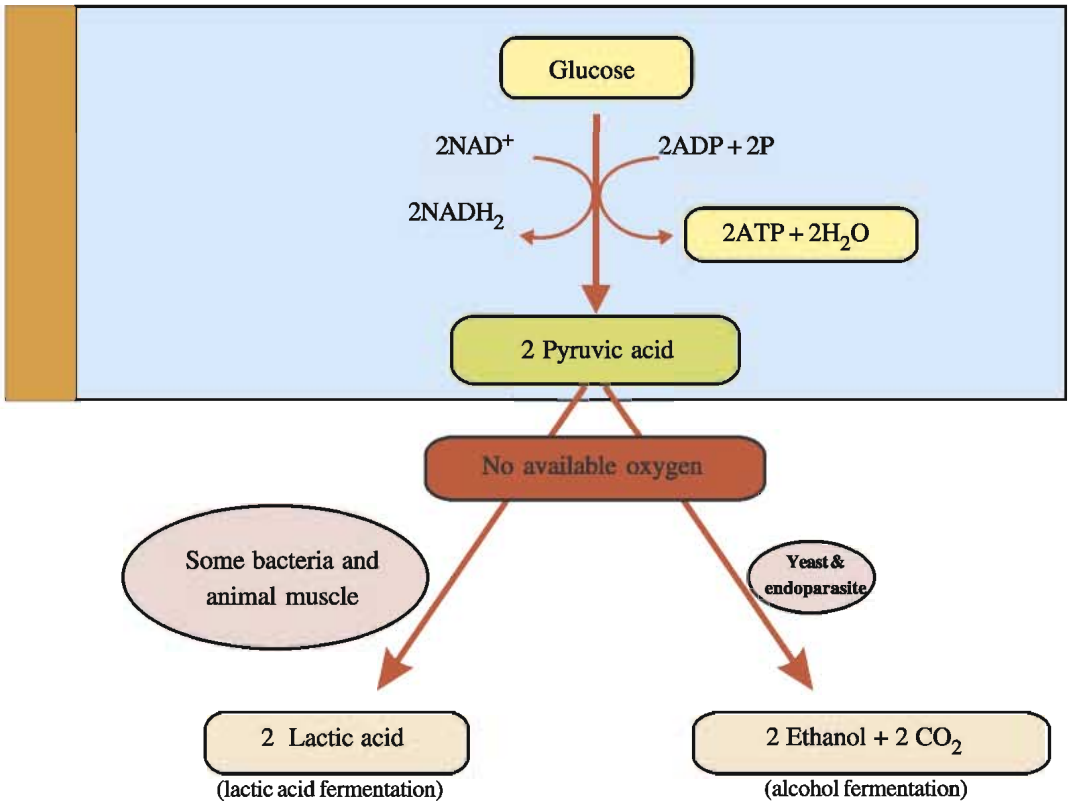
Ethyl alcohol

Lactic acid fermentation : In this type of fermentation, the pyruvic acid is converted into lactic acid by the enzyme lactate dehydrogenase and coenzyme $NADH_2$. The $NADH_2$ is oxidized to NAD.

$$2\ CH_3\ CO\ COOH + 2\ NADH_2 \longrightarrow 2\ CH_3\ CHOHCOOH + 2\ NAD$$

Lactic acid

Lactic acid fermentation takes place in the muscles of animals during exercise when oxygen is inadequate for cellular respiration.



Fermentation

In both, alcoholic and lactic acid fermentations less than seven percent of the energy in glucose is released and not all of it is trapped as high energy bonds of ATP. The end product is either lactic acid or alcohol.

Aerobic Respiration :

Aerobic respiration is the process that leads to a complete oxidation of glucose in presence of oxygen and releases CO_2, water and large amount of energy. Aerobic respiration includes Krebs cycle and oxidative phosphorylation in addition to glycolysis. At the end of glycolysis two molecules of pyruvic acid are produced, which enter the mitochondria wherein all the reactions of Krebs cycle and oxidative phosphorylation are carried out with the help of enzymes. It is interesting to note that Krebs cycle takes place in the matrix of mitochondria while oxidative phosphorylation takes place on the inner membrane of mitochondria. In mitochondria, the pyruvic acid having 3 carbon atoms undergoes complete oxidation releasing 3 molecules of CO_2. The presence of O_2 is inevitable during the entire process of Krebs cycle and oxidative phosphorylation.

Before entering into the Krebs cycle, pyruvic acid is decarboxylated and simultaneously it is also oxidized. As a result, one molecule of CO_2 is released and NAD is converted to $NADH_2$. The resulting 2C molecule is an acetate unit. Cofactor-A (CO.A) acting as a coenzyme, accepts this 2C unit and becomes acetyl co-enzyme-A. The entire reaction is catalyse by an enzyme pyruvate dehydrogenase.

$$\text{Pyruvic acid} + \text{CoA} + \text{NAD} \xrightarrow{\text{Pyruvate dehydrogenase}} \text{Acetyl Co.A} + CO_2 + \text{NAD.}H_2$$

The acetyl Co.A then enters a cyclic pathway, Tricarboxylic acid cycle, which is commonly called as Krebs cycle after the scientist Hans Krebs who first elucidated it. Acetyl Co-A is the intermediate compound linking glycolysis to Krebs' cycle.

Tricarboxylic Acid Cycle : All reactions of Krebs cycle are carried out in the matrix of mitochondrion.

Krebs cycle begins when a 4-C- organic acid molecule of oxaloacetic acid which reacts with 2C containing acetyl-co-enzyme-A. A 6-C acid called citric acid is formed as a result. Thus, as the first substance formed is citric acid, this cycle is also known as citric acid cycle.

Citric acid is converted into its isomer called - isocitric acid. Isocitric acid undergoes decarboxylation and dehydrogenation to form a 5-C α - Ketoglutaric acid. The $2H^+$ released are accepted by NAD which then becomes $NADH_2$. CO_2 is released.

α -Ketoglutaric acid undergoes decarboxylation and dehydrogenation to form a 4-C succinic acid. The $2H^+$ released are accepted by NAD which then becomes $NADH_2$. CO_2 is released.

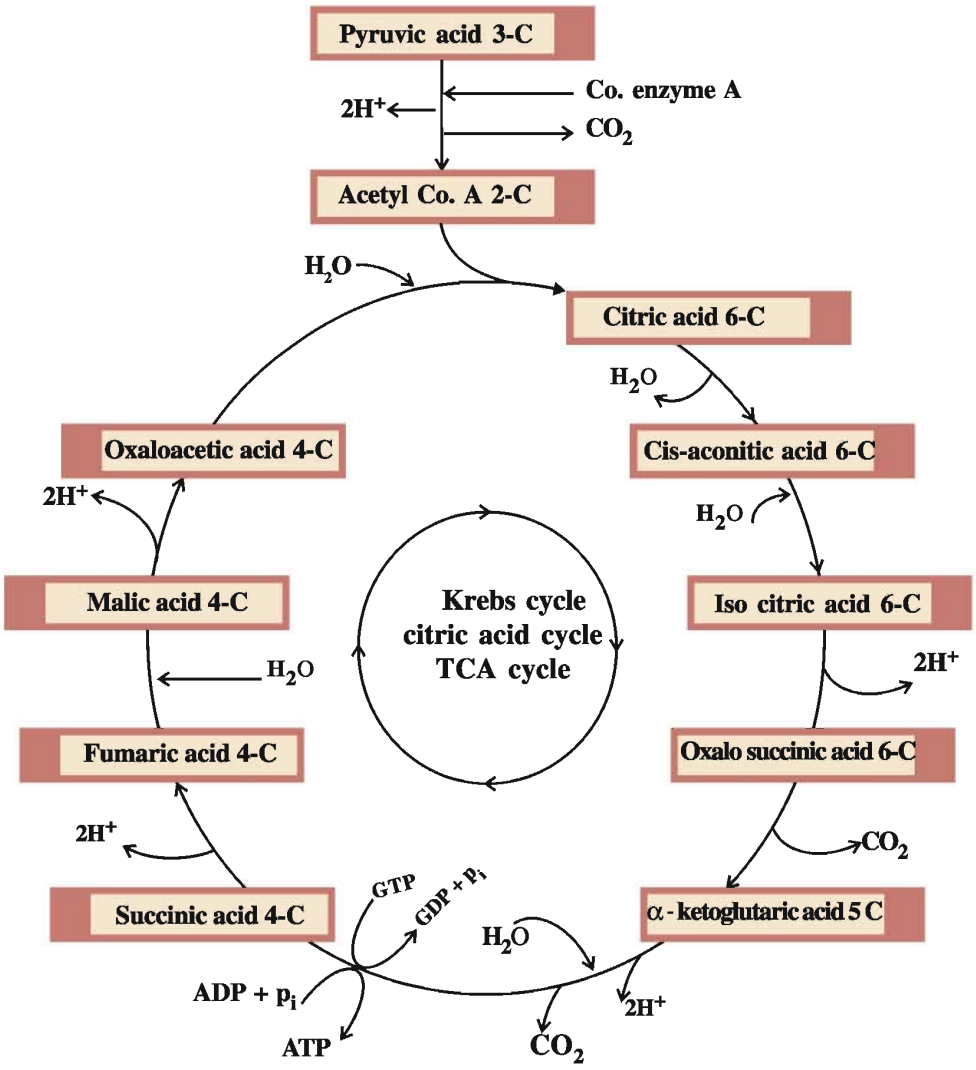
Succinic acid is dehydrogenated to form fumaric acid. The $2H^+$ released are accepted by FAD which then becomes $FADH_2$.

Fumaric acid receives one molecule of H_2O and is transformed into malic acid. Malic acid gets dehydrogenated and regenerates the oxaloacetic - acid. The released $2H^+$ are accepted by NAD which then becomes $NADH_2$.

Thus, when 1 molecule of pyruvic acid passes through the process of Krebs cycle, 3 molecules of CO_2 are released and at 5 different stages $2H^+$ and $2e^-$ are released.

When this occurs to both pyruvic acid molecules, a total of 6 CO_2 are released. Thus, the 6-C glucose molecule is completely decomposed. The summary equation for this phase of respiration may be written as follows:

$$\text{Pyruvic acid} + 4NAD^+ + FAD^+ + 3H_2O + ADP + Pi \longrightarrow 3CO_2 + 4NADH + 4H^+ + FADH_2 + ATP$$



Krebs Cycle

As, most of the organic acids, associated with this cycle, possess three carboxylic groups, this cycle is also known as TCA cycle (Tricarboxylic Acid Cycle).

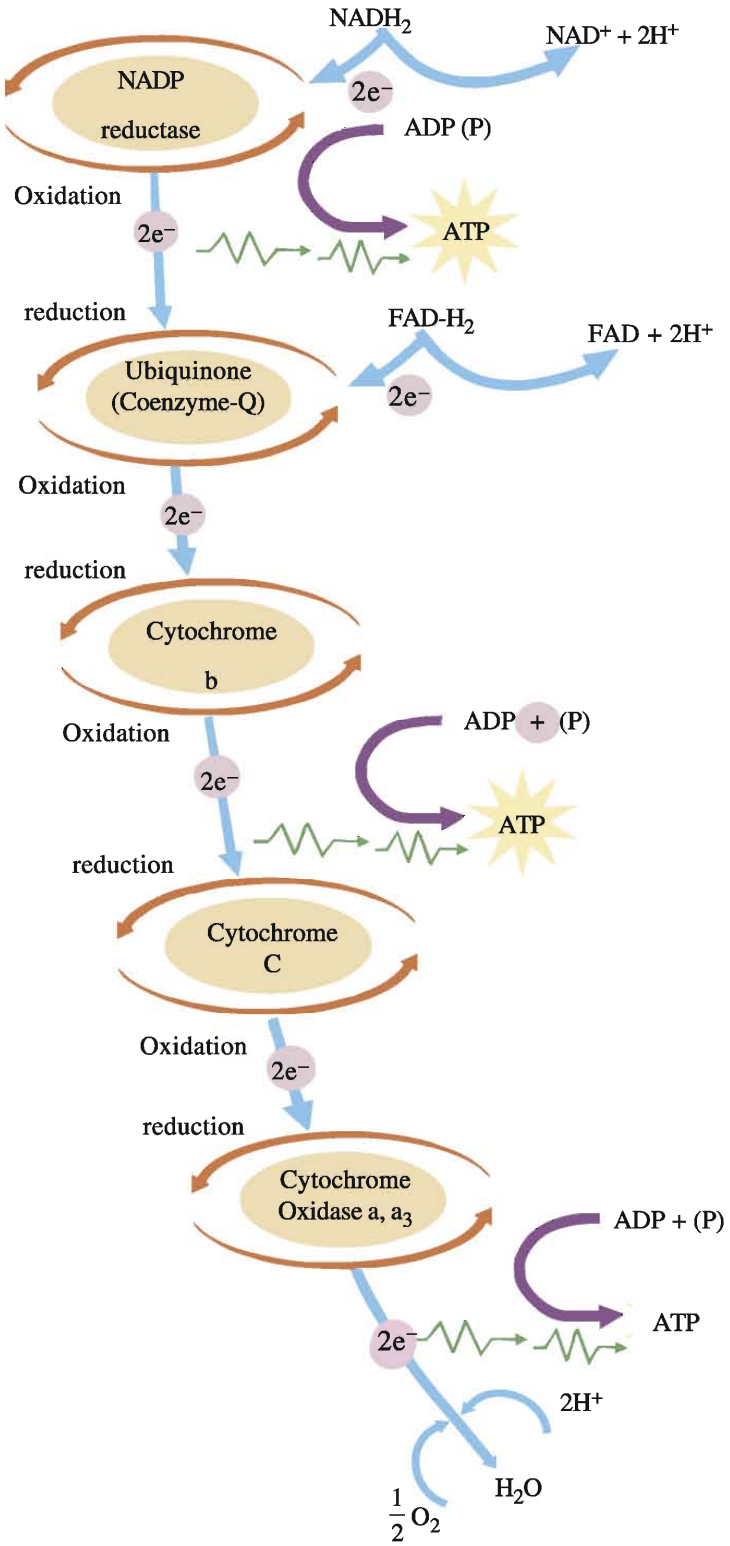
Significance of Krebs Cycle :

(1) It provides a pathway for complete breakdown of glucose.

(2) It provides the main pathway for synthesis of ATP.

(3) Various carbon - complexes formed during this cycle provide necessary components for growth and maintenance of cell. These components are utilized in the synthesis of substances like amino acids, nucleotides, fats, chlorophyll and cytochromes.

Electron Transport System (ETS) and Oxidative Phosphorylation : We observed that by the end of Krebs cycle, the molecule of glucose is completely degraded. However, unless the $NADH_2^-$ and $FADH_2$ formed during this process are transported to atmospheric O_2 and oxidized, no energy is released. Thus, transportation of $2H^+$ and $2e^-$ towards O_2 is essential. The metabolic pathway through which the electron passes from one carrier to another is called the electron transport system (ETS). This system is performed on inner membrane of mitochondria.



Oxidative Phosphorylation

The energy released during this process of oxidation is stored in the formation of ATP from ADP. As the phosphorylation of ADP to ATP occurs through the oxidation - energy, this phosphorylation is called oxidative-phosphorylation. This process takes place in units arranged on the cristae membranes of mitochondria.

First of all, $NADH_2$ transfers its $2H^+$ and $2e^-$ to NADP - an electron-acceptor substance. As a result NAD is released and NADP accepts $2H^+$ and $2e^-$. The $2H^+$ remain in the matrix and $2e^-$ are further transported.

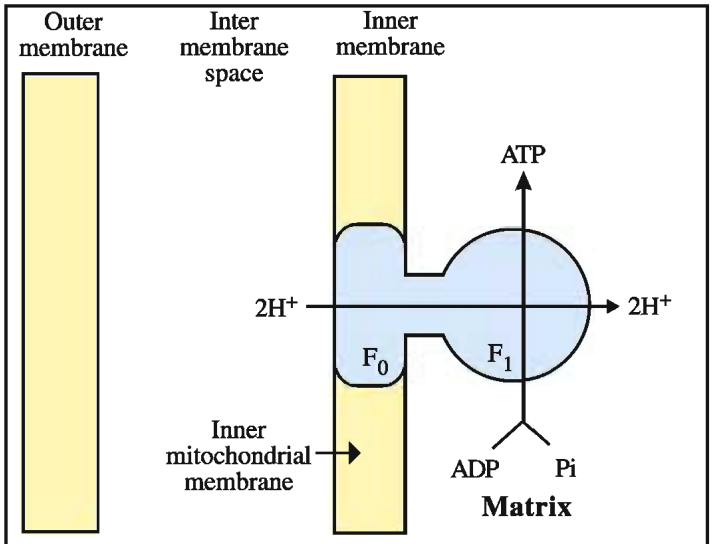
$2e^-$ from NADP enter the electron - carrier ubiquinone, from ubiquinone, they are further transported through a sequence of various cytochromes. The terminal acceptors in cytochrome chain are cytochromes *a* and a_3 *(cytochrome oxidase)*. These cytochromes collect $2H^+$ from the matrix and along with the $2e^-$ received by them combine with $\frac{1}{2}O_2$ obtained from the atmosphere and form H_2O.

The $2H^+$ and $2e^-$ from $FADH_2$ are also transported through ubiquinone (Coenzyme-a).

The energy released during transport of $2H^+$ from NAD to O_2, is stored in the synthesis of 3 ATP from ADP. Whereas during transport of $2H^+$ from FAD to O_2, energy released is stored in the synthesis of 2 ATP.

Mechanism of chemiosmotic generation of ATP :

You have already studied about the mechanism of membrane linked ATP synthesis as explained by chemiosmotic hypothesis in the chapter of photosyntheis. Since mitochondrial membrane is impermeable to protons, these cannot be diffused back into the matrix across the membrane. However, these can enter the membrane via a proton channel established by the membrane bound adenosine triphosphatase (ATPase). ATPase is a multienzyme complex containing two parts F_0 and F_1. F_0 component is embedded in the membrane forming a channel. Through this channel the protons flow to F_1. The F_1 headpiece is a peripheral membrane protein complex and contains the site for the synthesis of ATP from ADP. For each ATP produced, $2H^+$ passes through F_0 from the intermembrane space to the matrix down the electrochemical proton gradient. Thus for each pair of protons flowing back into matrix, one molecule of ATP is synthesized. So for three pairs of protons, three molecules of ATP are generated.



Diagramatic representation of ATP synthesis in mitochondria

Since $FADH_2$ transports only 2 pairs of protons outside the membrane through F_0 and F_1 complex, only two ATP molecules are produced.

The respiratory balance sheet

It is possible to calculate the net gain of ATP for every glucose molecule oxidized. The details of ATP synthesis during aerobic respiration can be described as under :

Synthesis of ATP during Glycolysis

Synthesis of substrate - based ATP – 2 ATP twice. (2ATP × 2 = 4 ATP)

Utilization during phosphorylation phase of glucose – 2 ATP.

Thus, as 4 ATP are formed and 2 ATP are utilized, there is a gain of 2 ATP.

During glycolysis, 2 molecules of $NADH_2$ are formed. Their oxidative phosphorylation will result in formation of 2 x 3 ATP = 6 ATP. Thus, during glycolysis synthesis of 8 ATP will occur.

Synthesis of ATP during Krebs Cycle : At four stages during degradation of pyruvic acid, $NADH_2$ is generated and at one stage $FADH_2$ is generated. During oxidative phosphorylation of 4 $NADH_2$ = 4 x 3 ATP = 12 ATP are formed and during phosphorylation of 1 $FADH_2$ = 1 x 2 ATP = 2 ATP are formed. Thus, a total of 14 ATP are synthesized.

As two molecules of pyrivic acid pass through this process a total of 28 (14 x 2 = 28 ATP) ATP are synthesized.

Moreover, during formation of succinic acid from, α Keto - glutaric acid, 1 ATP formation occurs which is substrate based. Thus, 2 ATP are formed. Thus total 30 ATP are formed during krebs cycle.

During aerobic respiration a total of 38 ATP are formed.

During glycolysis	:	6	through oxidative - phosphorylation
		2	through, substrate - based phosphorylation
During Krebs cycle	:	28	through oxidative - phosphorylation
		2	through substrate - based phosphorylation.
	Total :	38 ATP	

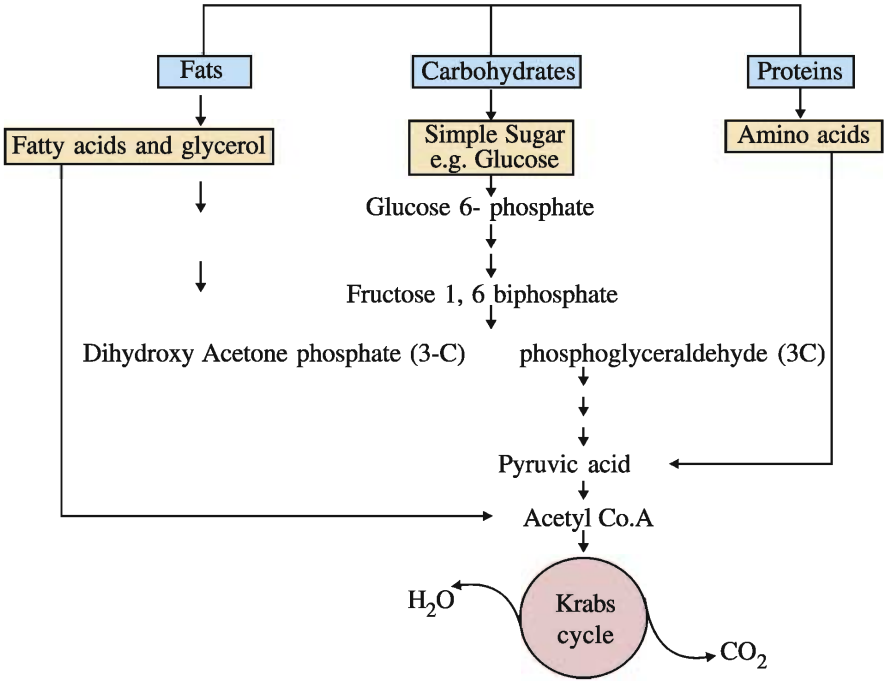
The efficiency of the process of transformation of potential chemical energy in glucose into the energy in ATP is about 45%. The remaining energy is dissipated as heat.

Moreover, as energy of two ATP is spent in transporting pyruvic acid into mitochondrion, the number of ATP is considered to be 36 in eukaryotic organisms.

Amphibolic Pathway

Carbohydrates are generally used as substrates for respiration. But before they enter respiration, they are converted into glucose. Other substrates can also be respired, but they do not enter the respiratory pathway at the first step. For example, fats need to be broken down into glycerol and fatty acids first. Now fatty acids first degraded into acetyl Co.A, which can enter into the respiratory pathway. Similarly, the glycerol is first converted into PGAL and then enter into respiratory pathway. In case of proteins, they are first broken down into amino acids which later converted in pyruvic acid and enter respiratory pathway.

The most important thing is that when fatty acids have to be respired, first they have to be broken down to acetyl Co.A which enters into respiratory pathway. But when organisms need to synthesise fatty acids, acetyl Co.A would be withdrawn from the respiratory pathway. Hence, the respiratory pathway comes into the picture both during breakdown and synthesis of fatty acids. Similarly, during breakdown and synthesis of proteins too. In this way respiratory pathway is involved in both anabolic and catabolic processes and hence it is also known as an amphibolic pathway rather than as a catabolic one.



Interrelationship among metabolic pathways showing respiration mediated breakdown of different organic molecules to CO_2 and H_2O

Respiratory Quotient

We know that during aerobic respiration O_2 is consumed and CO_2 is released. The ratio of released CO_2 to the O_2 consumed during respiration is called -Respiratory quotient - (RQ).

$$\text{Respiratory Quotient} = \frac{CO_2 \text{ released during respiration}}{O_2 \text{ consumed during respiration}}$$

The value of RQ depends on the material utilized for respiration. For example

Carbohydrates : For carbohydrats the value of RQ is 1. This would mean that the amount of O_2 utilized and the amount of CO_2 released are the same.

Equation : $C_6H_{12}O_6 + 6O_2 \longrightarrow 6CO_2 + 6H_2O + \text{Energy}$

$$RQ = \frac{6CO_2}{6O_2} = 1$$

Fats : For fats, the value of RQ is less than 1. This would mean that there is much less oxygen in the constituion of fat as compared to that in carbohydrates. Thus, they need more O_2 for respiration.

As an example, the equation for respiration of Tripalmitin is as under :

$2\ (C_{51}\ H_{98}\ O_6) + 145\ O_2 \longrightarrow 102\ CO_2 + 98\ H_2O + \text{Energy}$

$$RQ = \frac{102CO_2}{145O_2} = 0.7$$

When proteins are respiratory substrate, the ratio would be about 0.9.

It is important to know that in living organism respiratory substances are more than one and pure proteins or fats are never used as respiratory substrates.

Summary

A constant supply of energy is essential to carry out physiological processes in living organisms. Based on the energy exchange there are two types of biological processes : (1) Endergonic processes and (2) exergonic processes. The breakdown of C-C bonds of complex compounds through oxidation within the cells releasing considerable amount of energy is called respiration. The substances which are oxidized during this process are known as respiratory substrates.

The breakdown of glucose to pyruvic acid is called glycolysis. Glycolysis occurs in the cytoplasm matrix of cells. During glycolysis glucose is converted into two molecules of pyruvic acids. There are three major ways in which different cells utilized pyruvic acid produced during glycolysis. These are lactic acid fermentation, alcoholic fermentation and aerobic respiration.

The end products of alcoholic fermentation is ethyl alcohol and CO_2 while that of lactic acid fermentation is lactic acid.

Aerobic reaspiration is the process that leads to a complete oxidation of glucose in presence of oxygen and releases CO_2, water and large amount of energy. Aerobic respiration includes Krebs cycle and oxidative phosphorylation in addition to glycolysis.

Before entering into the Krebs cycle, pyruvic acid is decarboxylated and simultaneously it is also oxidized producing Acetyl Co-A, CO_2 and $NADH_2$.

All reactions of Krebs cycle are carried out in the matrix of mitochondrion.

The metabolic pathway through which the electron passes from one carrier to another is called the electron transport system (ETS). This system is located on inner membrane of mitochondria.

During electron transport system, energy released during transport of H_2 from NAD to O_2, is stored in the synthesis of 3 ATP from ADP. Whereas during transport of H_2 from FAD to O_2, energy released is stored in the synthesis of 2 ATP from ADP.

Respiratory pathway is involved in both anabolic and catabolic processes and hence it is also known as an amphibolic pathway rather than as a catabolic one.

During aerobic respiration, O_2 is consumed and CO_2 is released. The ratio of released CO_2 to the O_2 consumed during respiration is called Respiratory quotient (RQ).

Exercise

1. Put a dark colour in a given circle for correct answer :

(1) TCA cycle occurs in...

(a) Mitochondria ○ (b) Chloroplast ○

(c) Cytoplasm ○ (d) Peroxisome ○

(2) Anaerobic process after glycolysis is...

(a) ETS ○ (b) Calvin cycle ○

(c) Krebs cycle ○ (d) None of these ○

(3) The end products of fermentation are...

(a) O_2 and acetaldehyde ○ (b) O_2 and ethyl alcohol ○

(c) CO_2 and ethyl alcohol ○ (d) CO_2 and acetaldehyde ○

(4) Electron transport system in mitochondria is located on...

(a) Outer membrane ○ (b) Inter cristae space ○

(c) Inner membrane ○ (d) Inner membrane space ○

(5) During fermentation in Yeast cell, alcohol is fermented from...

(a) Protein ○ (b) Lipid ○

(c) Sugar ○ (d) Nucleic acids ○

(6) The key intermediate compound linking glycolysis to the Krebs cycle is...

(a) Malic acid ○ (b) Pyruvic acid ○

(c) Succinic acid ○ (d) Acetyl Co-A ○

(7) In Krebs cycle, FAD participates as electron acceptor during the conversion of...

(a) Succinyl Co.A to succinic acid ○ (b) Fumaric acid to malic acid ○

(c) Succinic acid to fumaric acid ○ (d) Malic acid to oxaloacetic acid ○

(8) Glycolysis takes place in...

(a) Mitochondria ○ (b) Cristae of mitochondria ○

(c) Cytoplasmic matrix ○ (d) Inner membrane of mitochondria ○

(9) Enzyme responsible for phosphorylation of glucose into glucose-6-phosphate is...

(a) ATPase ○ (b) Hexokinase ○

(c) Dehydrogenase ○ (d) Oxidase ○

(10) During glycolysis one molecule of glucose produces two molecules of...

(a) Acetyl Co-A ○ (b) Pyruvic acid ○

(c) Acetaldehyde ○ (d) Malic acid. ○

2. Answer the following questions in short :

(1) Why is Krebs cycle called TCA cycle ?

(2) Define cellular respiration, glycolysis.

(3) Define endergonic and exergonic processes.

(4) Mention the sites of Glycolysis, Krebs cycle and Oxidative phosphorylation

(5) Give the chemical equation of respiration

(6) Give chemical equation of Kreb's cycle.

(7) Define anaerobic respiration.

(8) What is electron transport system ?

(9) In eukaryotic organisms, how many net ATP molecules are produced from oxidation of one glucose molecule .

3. Do as directed :

(1) Differentiated between aerobic and anaerobic respiration.

(2) Describe respiratory quotient.

(3) What is significance of Krebs cycle ?

(4) Describe lactic acid fermentation ?

(5) Describe alcoholic fermentation ?

4. Answer the following questions in detail :

(1) Write, in detail, whatever you known about glycolysis.

(2) Describe Krebs cycle.

(3) Explain different steps involved in oxidative phosphorylation.

(4) Explain electron transport system.

(5) Sketch the diagram of Krebs cycle.