CHAPTER -14 RESPIRATION IN PLANTS

Respiration is a process during which the food materials are broken down into CO₂ and water with the release of energy contained in them for the synthesis of ATP molecules.

ATP (Adenosine Triphosphate) is broken down when ever and where ever energy needs to be utilized, hence ATP acts as the "energy currency of the cell".

♦ $C_6H_{12}O_6 + 6O_2 \rightarrow 6CO_2 + 6H_2O + Energy [2870KJ]$

It is an -

- ◆ Oxidative (addition of O₂)
- Exergonic (energy releasing) &
- ♦ Catabolic (destructive) process

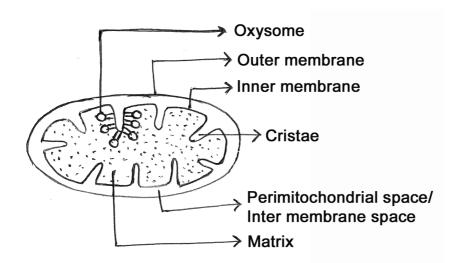
Some scientists prefer to call respiration as an **Amphibolic process** [ie., both anabolic and catabolic] because some of the respiratory intermediates are used in the synthesis of certain biomolecules like glutamic acid, Aspartic acid etc.

• In eukaryotes, the respiratory process takes place in the :

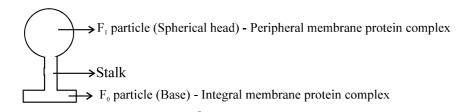
<u>Cytoplasm</u> (Glycolysis) and in the <u>Mitochondria</u> (Krebs cycle & ETC)

(ETC - Electron Transport Chain)

- ♦ The cell organelle concerned with respiration is Mitochondria, so it is also known as -
 - ♦ Power house of the cell
 - ♦ Cell battery
 - ♦ Cell furnace
 - ♦ Biological furnace
 - ♦ ATP factory



- Mitochondria is bounded by a double membrane. The outer membrane is smooth while the inner membrane has many infoldings called cristae. The space in between the two membranes is called peri-mitochondrial space or intermembrane space. The inner membrane encloses a space, filled with matrix, The matrix contains DNA, RNA, ribosomes of 70S type and various enzymes concerned with Krebs cycle.
- ◆ The electron microscopic studies have revealed that certain tennis racket like structures are present on the cristae with a base (F₀ - particle), stalk and a spherical head (F₁ particle) called oxysomes / Fernandez - Moran particle (FM particles), / F₀ - F₁ particles / Elementary particles.



Oxysome

- ◆ The F₁ head piece is a peripheral membrane protein complex and contains the site for synthesis of ATP from ADP and inorganic phosphate during oxidative phosphorylation.
- ◆ F₀ is an integral membrane protein complex that forms the channel through which protons cross the inner membrane.
- ♦ In **Prokaryotic organisms** like bacteria, the respiratory enzymes are found in the **plasma** membrane / cell membrane, the infolding of which are called **Mesosomes / Chondrioids**.

Respiratory Substrates

The compounds that are broken down during respiration are called respiratory substrates. They are generally **carbohydrates**, **fats** and **proteins**. Usually carbohydrates are oxidised to release energy, but fats, proteins and even organic acids can be used as respiratory substrate in some plants under certain conditions. Proteins are used up only when carbohydrates and fats are not available. When carbohydrates, fats and proteins are not available, organic acids are used as respiratory substrates.

Floating respiration (Common type)

It is the respiration in which carbohydrates or fats are used as respiratory substrates.

Protoplasmic respiration

The respiration in which **proteins** are used as respiratory substrate.

♦ Salt respiration

Increased rate of respiration exhibited by plants during **active absorption** (absorption against concentration gradient)

♦ Photorespiration

Respiration occurring in photosynthetic tissues in which there is no release of energy. The substrate is a 2 carbon glycolate, hence photorespiration is also called **Glycolate cycle / C_2 cycle**. The cell organelles concerned with photorespiration are **chloroplast**, **peroxisome** and **mitochondria**.

♦ Respiratory Climacterics

Certain fruits like Apple, Mango etc. exhibit an increased rate of respiration at the time of ripening of fruits, referred to as respiratory climacerics. The hormone responsible for this is ethylene (only gaseous hormone) so it is also known as climacteric hormone.

♦ Anti-transpirant

Any substance which can reduce the rate of transpiration.

eg. ABA (Abscisic acid) → Natural Anti-transpirant

Respiratory Quotient [R.Q] / Respiratory ratio

The ratio of the volume of CO_2 evolved to the volume of O_2 consumed in respiration is called respiratory quotient / Respiratory ratio.

$$R.Q = \frac{\text{Volume of } CO_2 \text{ evolved}}{\text{Volume of } O_2 \text{ consumed}}$$

- ◆ The R.Q. depends upon the **type of respiratory substrate** used during respiration.
 - 1) Carbohydrates RQ = One / Unity
 - 2) Fats or proteins Less than one (<1)
 - 3) Organic acids Greater than one (>1)
 - 4) Succulent xerophytes / CAM plants R.Q = Zero
 - 5) Anaerobic respiration R.Q Infinity (∞)

♦ When carbohydrates are used as respiratory substrate, the volume of CO₂ evolved and O₂ consumed are equal, so that the R.Q value is one / unity.

eg.
$$\frac{6\text{CO}_2}{6\text{O}_2} = 1$$

◆ Fats require more O₂ than the amount of CO₂ released, so that the R.Q value is less than one.

Eg. Tripalmitin (a fat) R.Q =
$$\frac{102CO_2}{145O_2} = 0.7$$

- ◆ Proteins also require more O₂ than the amount of CO₂ released, so that the R.Q value is less than one often upto 0.9
- ◆ Organic acids are rich in oxygen, so it require lesser O₂ for their oxidation, so that R.Q value is greater than one.

eg. Tartaric acid - R.Q =
$$\frac{8\text{CO}_2}{5\text{O}_2}$$
 = 1.6

Oxalic acid - R.Q =
$$\frac{4\text{CO}_2}{1\text{ O}_2}$$
 = 4

Malic acid - RQ =
$$\frac{4\text{CO}_2}{3\text{O}_2} = 1.33$$

♦ In succulent xerophytes / CAM plants, the RQ value is zero.

(These plants possess scotoactive stomata ie, the stomata which are opening during night and closed during day time.)

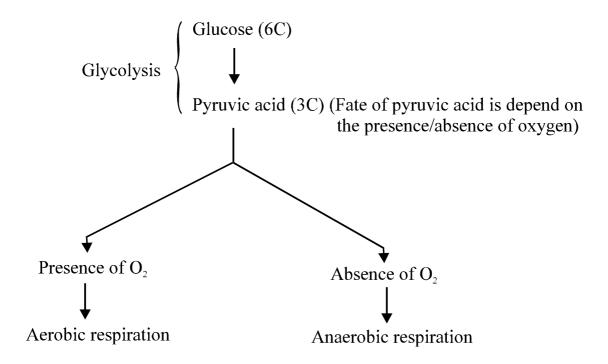
In such plants, during night, the food materials are incompletely broken down ie, not reached upto CO_2 release. During day time, food materials are completely broken down (reached upto CO_2 release) but at that time their stomata were closed. The CO_2 evolved is simultaneously used up in the synthesis of sugars.

So that there is no release of CO₂ either at night or at day. So that the R.Q value is zero.

♦ In Anaerobic respiration, RQ value is infinity (∞) because CO_2 is evolved but O_2 is not used.

eg.
$$\frac{2\text{CO}_2}{0\text{O}_2} = \infty$$

Difference between aerobic and anaerobic respiration



Aerobic Respiration	Anaerobic respiration
It is the common type of respiration	It occurs in yeast and some bacteria
Food materials are completely oxidised (2870KJ)	Food materials are incompletely oxidised (247 KJ)
Site of reaction is cytoplasm & mitochondria	Site of reaction is cytoplasm
End products are CO ₂ , H ₂ O and 38ATP molecules	End products are Ethanol, CO ₂ and 2ATP or Lactic acid and 2ATP
It involves Glycolysis, Krebs cycle and ETS	Glycolysis is the only process and fermentation is a type of anaerobic respiration

Aerobic Respiration

The common pathway of aerobic respiration occurs in three phases namely:

- 1) Glycolysis Site is cytoplasm (agranular part of which is called **cytosol**)
- 2) Krebs cycle site is mitochondrial matrix
- 3) ETS and Oxidative phosphorylation site is inner mitochondrial membrane.

Glycolysis

The breakdown of glucose to pyruvic acid, which occur in the agranular part of cytoplasm ie, cytosol is called glycolysis.

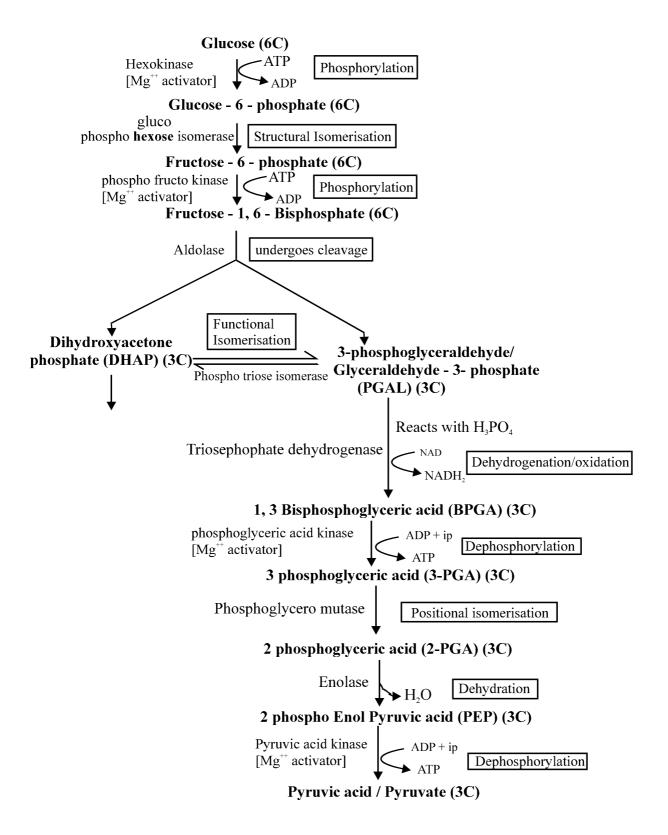
The term 'Glycolysis' was originated from two greek words.

They are:

Glycos = sugar

Lysis = splitting

- ◆ The details regarding Glycolysis was revealed by the work of 3 scientists Gustav Embden, Otto Meyerhof and J. Parnas so the name **EMP pathway**.
- ♦ It is also known as **common respiratory pathway** because, upto the formation of pyruvic acid, it is common for both aerobic and anaerobic respiration.
- ♦ Also known as **cytoplasmic respiration** because, the site of glycolysis is cytoplasm.
- Known by the name, Triosis, because during glycolysis, hexose (6C), ie, glucose is splitted to form trioses (3C)



- ◆ During Glycolysis, there is no need of any cell organelle, no use of O₂, no release of CO₂ (ie, no decarboxylation), whether O₂ is present or not is unconcerned.
- Pyruvic acid is the key product of glycolysis and is the common intermediate of both aerobic and anaerobic respiration.
- The trioses formed during glycolysis are DHAP and PGAL
- ♦ The step in which oxidation occurs in glycolysis is PGAL →BPGA
- ♦ The steps in which utilization of ATP occurs :
 - *Glucose → Glucose 6 phosphate
 - * Fructose-6-phosphate → Fructose 1, 6-Bisphosphate
- ♦ The steps in which synthesis of ATP occurs :
 - *1, 3 Bisphosphoglyceric acid (BPGA) → 3-phosphoglyceric acid (3-PGA)
 - *2-phosphoenol pyruvic acid (PEP) → Pyruvic acid
- ◆ The mineral needed as an enzyme activator is Mg⁺⁺

♦ Net gain of Glycolysis

- ♦ 2 molecules of pyruvic acid
- ♦ 2 molecules of NADH₂ (1NADH₂ \rightarrow 3ATP; \therefore 2 × 3 = 6ATP)
- ♦ 2 molecules of ATP

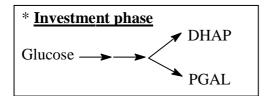
(4 ATP formed from 2 branches through substrate phosphorylation and 2 ATP used up)

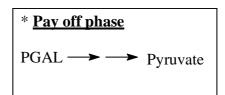
 \therefore Total ATP = 6 + 2 = 8ATP (ie, from 2NADH₂ + 2ATP)

♦ Substrate phosphorylation

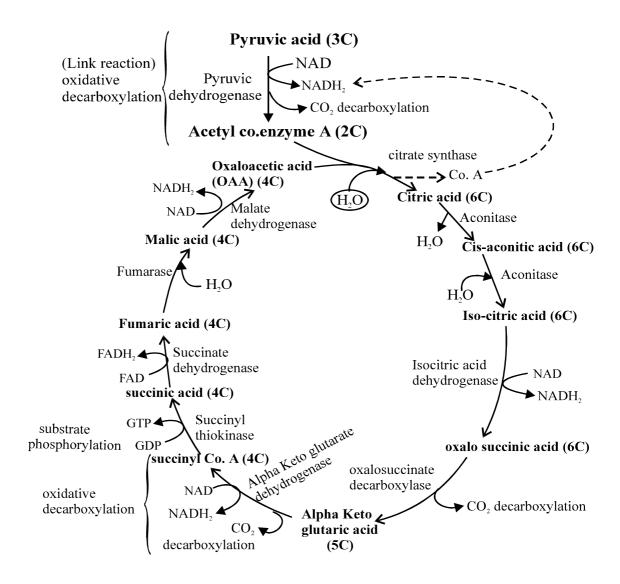
Direct ATP production without the involvement of Electron Transport System (ETS).

eg.
$$ADP + ip \Rightarrow ATP$$





KREBS CYCLE



- ◆ NAD Nicotinamide Adenine Dinucleotide [universal / common hydrogen acceptor]
- ♦ FAD Flavin Adenine Dinucleotide
- ♦ GDP Guanosine Diphosphate
- ◆ GTP Guanosine Triphosphate [High energy phosphate in Krebs cycle]
- ♦ Krebs cycle (after the Scientist Hans Adolf Krebs) is also known as -
 - 1. Citric acid cycle because first stable product in Krebs cycle is citric acid.
 - **2. Tricarboxylic acid cycle (TCA cycle)** because citric acid is a tricarboxylic acid (ie., having 3-COOH group)

- 3. Organic acid cycle because many organic acids are formed during Krebs cycle.
- 4. Mitochondrial respiration because the site of Krebs cycle is mitochondria.
 - * All 6C compounds in Krebs cycle ie, Citric acid, cis-aconitic acid, isocitric acid and oxalosuccinic acid) are **Tricarboxylic** (ie, having 3 COOH)
 - * Succinyl Co. A is **Monocarboxylic** (ie, having 1 COOH)
 - * All other 4C compounds and 5C compound ie, succinic acid, fumaric acid, Malic acid, oxalo acetic acid & Alpha-ketoglutaric acid) are **Dicarboxylic** (ie having 2 COOH)
- During aerobic respiration, the 3C pyruvic acid (it enters the mitochondrial matrix), formed as a result of glycolysis is not directly participate in the second phase of aerobic respiration ie, Krebs cycle.
- ♦ By **oxidative decarboxylation** (ie, both oxidation / dehydrogenation and decarboxylation) the 3C pyruvic acid is converted to 2C Acetyl co. enzyme A. [Link reaction between glycolysis and Krebs cycle or Gate way reaction]
- ◆ A multi-enzyme complex (ie pyruvate dehydrogenase) and 5 co-factors are involved during this conversion. They are
 - (1) Lipoic acid
 - (2) NAD
 - $(3) Mg^{++}$
 - (4) Co. enzyme A
 - (5) TPP (Thiamine Pyro Phosphate)
- ♦ Krebs cycle begins with the reaction between 2C acetyl Co. A and 4C oxalo acetic acid, resulting in the formation of 6C citric acid.
- ♦ A single turn of Krebs cycle yields
 - \rightarrow 3 molecules of NADH₂ (1 NADH₂ \rightarrow 3ATP, \therefore 3 x 3 = 9 ATP)
 - \rightarrow 1 FADH₂ (1 FADH₂ \rightarrow 2ATP; \therefore 1 × 2 = 2 ATP)
 - \rightarrow 1 GTP (1 GTP = 1ATP; \therefore 1 × 1 = 1 ATP)
 - ∴ Total ATP = 9 + 2 + 1 = 12 ATP
 - ◆ Total number of CO₂ evolved during oxidation of one molecule of glucose 6CO₂
 Hint: (4 molecules of CO₂ from 2 Krebs cycles and 2 CO₂ through 2 link reactions)
 - ◆ There are 4 reduced co-enzymes (ie, 3 NADH₂ and 1 FADH₂) in a Krebs cycle. In mitochondrial matrix, there are 5 reduced co-enzymes.

♦ In a Krebs cycle there are :

- ◆ 4 oxidations / dehydrogenations (ie, 3NADH₂ and 1 FADH₂)
- ♦ 2 decarboxylations and
- ◆ 1 substrate level phosphorylation.
- ◆ The GTP produced during the conversion of succinyl co. A to succinic acid, later transfer its phosphate group to ADP to form a molecule of ATP, without the involvement of ETS, so the process involved here is substrate phosphorylation.

♦ Acetyl Co. A

- ♦ Connecting link between glycolysis and Krebs cycle
- Only 2C compound formed in aerobic respiration.
- ♦ First entrant in Krebs cycle and formed in matrix due to oxidative decarboxylation of pyruvate.
- Oxalo acetic acid Final 4C compound in Krebs cycle and the first member in Krebs cycle.
- Citric acid is the first stable product in Krebs cycle.
- ◆ AlphakKetoglutaric acid Only 5C compound in Krebs cycle.
- ◆ Succinyl Co.A is the first formed 4 carbon compound and only monocarboxylic compound in Krebs cycle.
- ◆ **Succinate dehydrogenase** is the enzyme present in the inner mitochondrial membrane. (All other enzymes of Krebs cycle are in the matrix of mitochondria).

Amphibolic steps

- 1) Pyruvic acid (3C) Acetyl Co. A (2C)
 - (Acetyl co. A is the precursor of Gibberellin)
- 2) Oxalo Succinic acid (6C) \rightarrow Ketoglutaric acid (5C)

(Ketoglutarate is involved in reductive amination)

3) Ketoglutaric acid (5C) Succinyl Co. A (4C)

(Succinyl Co. A involved in the synthesis of chlorophyll)

ATP Calculation

- 1) During Glycolysis, when PGAL is converted to BPGA
- 1NADH₂

- Dehydrogenation / oxidation during glycolysis
- 2) Pyruvic acid Acetyl Co. A

- 1NADH₂
- First oxidative decarboxylation in Respiration
 - Link reaction / Gateway reaction
- 3) Isocitric acid Oxalosuccinic acid

- 1NADH₂

[*1st oxidation / dehydrogenation in Krebs cycle]

4) Ketoglutaric acid Succinyl Co.A - 1 NADH₂ [*Only step in Krebs cycle, where oxidative decarboxylation occurs Second oxidative decarboxylation during respiration [™]1st or last oxidative decarboxylation in Krebs cycle Final decarboxylation in Krebs cycle or respiration ^ℊ 2nd oxidation in Krebs cycle Final decarboxylation in Krebs cycle 5) Malic acid Oxalo acetic acid - 1NADH₂ [Final dehydrogenation / oxidation in Krebs cycle Total - 5 NADH₂ From 1 NADH₂ → 3ATP molecules \therefore from 5NADH₂ \rightarrow 5 × 3 = **15ATP** 6) Succinic acid Fumaric acid - 1FADH₂ [*Only step in Krebs cycle where FAD is involved 3rd oxidation / dehydrogenation in Krebs cycle] From 1 FADH₂ \rightarrow 2 ATP molecules. \therefore from 1FADH₂ \rightarrow 1 x 2 = **2 ATP** 7) Succinyl Co. A Succinic acid - 1GTP [Substrate phosphorylation in Krebs cycle] 1 GTP = 1 ATP; $\therefore 1 \times 1 = 1 \text{ ATP}$ Total ATP = 15 + 2 + 1 = 18 ATP ♦ From 2 branches ie, from DHAP and PGAL - 18 x 2 = 36 ATP Net gain of ATP from Glycolysis = 2 ATP Grand total = 36 + 2 = 38 ATP

- Qn. Total number of co-enzymes (ie, NADH₂ & FADH₂) reduced during the complete oxidation of one molecule of Glucose?
- Ans. 12 [10 NADH₂ & 2 FADH₂]

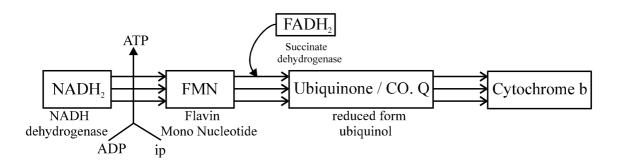
Hint:

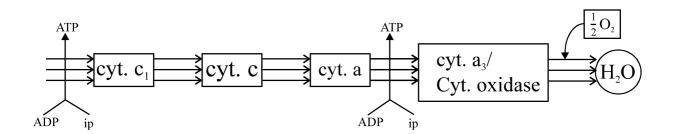
- * 2NADH₂ from Glycolysis ie, in cytosol
- * 2NADH₂ from 2 Link reactions * 6NADH₂ from 2 Krebs cycles * 2FADH₂ from 2 Krebs cycles Mitochondrial matrix

Total 12 co-enzymes

(Out of 12, 10 molecules are formed in mitochondria and 2 in cytoplasm ie, in cytosol)

Electron Transport System [ETS] and Oxidative Phosphorylation





- ◆ The reduced co-enzymes NADH₂ and FADH₂ are oxidised by the help of atmospheric oxygen in a step-wise electron transfer.
- ◆ For this the electrons from the reduced co-enzymes are released and transported step wise through intermediate electron carriers like FMN, Ubiquinone (reduced form is ubiquinol) and different cytochromes such as b, c₁, c, a and a₃. This is called Electron Transport System (ETS) and it forms an Electron Transport Chain [ETC], which is operated in the inner mitochondrial membrane.

◆ Electron carriers (Total - 7)

- 1. FMN
- 2) Ubiquinone / co.enzyme Q Mobile carrier between complex II and III

5) Cytochrome c - Mobile carrier between complex III and IV

* Complex I	NADH dehydrogenase
* Complex II	FADH ₂ / Succinate dehydrogenase
* Complex III	Cytochrome bc ₁ complex
* Complex IV	Cytochrome c oxidase complex containing cyt. a, a ₃ & 2 copper centres
* Complex V	ATP synthase (major components are F_0 and F_1)

- ◆ Electrons from NADH₂ are oxidised by an NADH dehydrogenase (complex 1) and electrons are then transferred to ubiquinone through FMN.
- Ubiquinone also receives reducing equivalents via, FADH₂ (complex II), that is generated during oxidation of succinate, through the activity of the enzyme succinate dehydrogenase (it is present in the inner mitochondrial membrane); in the Citric acid cycle ie, Krebs cycle.
- ♦ When protons move from F₀ particle to F₁ particle, it bring about configurational changes in the F₁ particle, so that F₁ particle acts as ATP synthase (Complex V), aiding in synthesis of ATP.
- ◆ The electron finally reacts with molecular oxygen ie, 1/2 O₂ taken from the atmosphere and proton H⁺ released during the electron transport chain resulting in the production of water.
- ♦ The last step of electron transport chain where the electrons along with protons from hydrogen are transferred to molecular oxygen is called **Terminal oxidation**.
- ◆ Simultaneously with the oxidation by electron transport, the energy contained in NADH₂ and FADH₂ are released and a part of it is used in production of ATP.

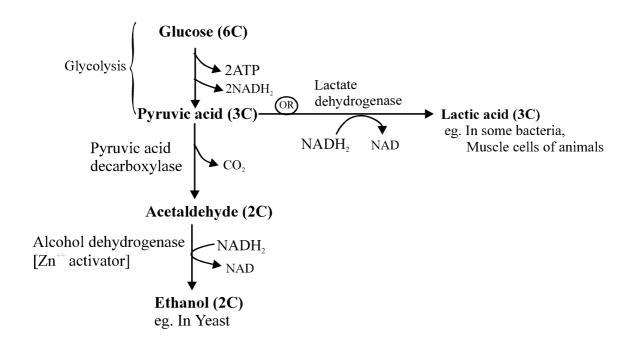
- The transport of electrons through the electron transport chain helps in synthesizing. ATP by coupling of ADP and inorganic phosphate (ip). This synthesis of ATP during aerobic respiration is called oxidative phosphorylation.
- The number of ATP molecules synthesised depends on the nature of the electron donor. Oxidation of one molecule of NADH₂ gives 3 molecules of ATP while that of one molecule of FADH₃ produces 2 molecules of ATP.
- ◆ The terminal donor of electrons to oxygen (final electron acceptor) for the formation of water in oxidative phosphorylation is from reduced cytochrome a₃ or cytochrome oxidase.
 - (**Cytochrome a**₃ it contain copper in addition to iron, With iron it pickup electrons and through copper it hands over electrons to O₂)
- ◆ Cytochrome c It is a small protein attached to outer surface of the inner membrane of mitochondria and acts as a mobile carrier of electrons between complex III and complex IV.

The Respiratory Balance Sheet

Calculations of the net gain of ATP for every glucose molecule oxidised is made on certain assumptions. They are ;

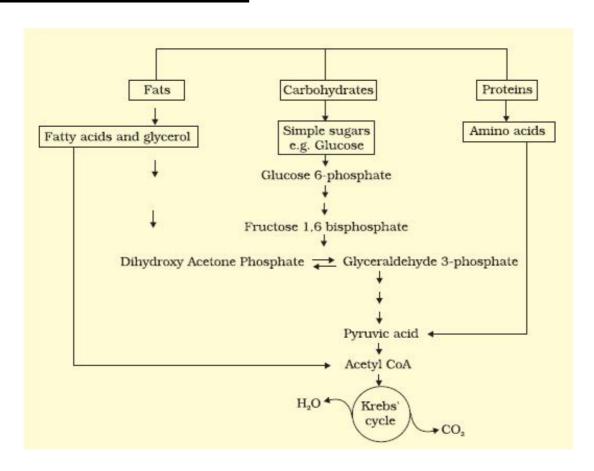
- ♦ There is a sequential, orderly pathway functioning, with one substrate forming the next and with glycolysis, TCA cycle and ETS pathway following one after another.
- ♦ The NADH synthesised in glycolysis is transferred into the mitochondria and undergoes oxidative phosphorylation.
- None of the intermediates in the pathway are utilised to synthesis any other compound.
- Only glucose is being respired no other alternative substrate are entering in the pathway at any of the intermediary stages.

Anaerobic Respiration



- When O₂ is limiting, NADH₂ and pyruvic acid formed as a result of glycolysis, begin to accumulate.
- Under this condition plants carry out fermentation ie, anaerobic respiration, leading to the formation of ethanol and CO₂ in alcoholic fermentation or Lactic acid in Lactic acid fermentation.
- In anaerobic respiration, the NADH₂ produced during glycolysis is used for the synthesis of Ethanol / Lactic acid. So that the net gain of ATP will be two which is directly formed through glycolysis.
- So the end products are ethanol, CO₂ and 2ATP or Lactic acid and 2 ATP.
- The site of reaction is cytoplasm.
- ◆ RQ in anaerobic respiration is **infinity** (CO₂ is formed but O₂ is not used). But in the case of Lactic acid fermentation, the RQ is unpredictable. (CO₂ is not formed and O₂ is not used)
- When one molecule of pyruvic acid is participating in fermentation, there is a loss of 3ATP. (formation of ethanol or Lactic acid utilizes one molecule of NADH₂ which is equivalent to 3ATP)

Fates of Different Substrates:



Interrelationship among metabolic pathways showing respiration mediated breakdown of different organic molecules to ${\rm CO_2}$ and ${\rm H_2O}$

- Glucose is the favoured substrate for respiration.
- ♦ All carbohydrates are usually first converted into glucose before they are used for respiration.
- ◆ Sucrose is converted into glucose and fructose by the enzyme, **invertase**, and these two monosaccharides can readily enter the glycolytic path way.
- ♦ Other substrates (fats & proteins) can also be respired, but they do not enter the respiratory pathway at the first step and can never used purely as substrates.
- ◆ Fats would need to be broken down into glycerol and fatty acids first; if fatty acids were to be respired they would first be degraded to acetyl Co. A and enter the pathway; glycerol would enter the pathway after being converted to PGAL (Phosphoglyceraldehyde).
- ♦ The proteins would be degraded by proteases, and the individual aminoacids (after deamination) depending on their structure would enter the pathway at some stage within the Kreb's cycle or even as pyruvate or acetyl Co.A.
- Since, some of the respiratory intermediates are used in different anabolic pathways to form important biomolecules like glutamic acid, aspartic acid etc., it would hence be better to consider the respiratory pathway as an **amphibolic pathway** rather than as a catabolic one.
- ◆ The metabolite which is common to respiration mediated breakdown of fats, carbohydrates and proteins is **Acetyl Co. enzyme A.**

Pentose Phosphate Pathway [PPP] / Hexose - Monophosphate Shunt / Direct oxidation of Glucose.

- It is an alternate pathway of aerobic respiration without glycolytic breakdown and Krebs cycle, where NADPH₂ is involved.
- ♦ It is activated when glycolysis is inhibited by any reason like poison.
- First reported by Warburg and Dickens. So the name Warburg Dicken's Cycle.
- ♦ The site of reaction is cytoplasm.

Factors affecting Respiration

- 1. **Temperature -** If temperature increases, enzymes denatured and if temperature decreases, enzymes inactivated.
- **2.** Oxygen In presence of O_2 aerobic respiration and in absence of O_2 anaerobic respiration.
- 3. CO₂ If conc. of O₂ increases, the rate of respiration decreases, because high conc. of CO₂ stimulates ABA and it closes stomata which reduces the uptake of oxygen.

5. Light - Rate of respiration increases with increase in light intensity.

- 6. Injury, wounds and Diseases enhances the rate of respiration.
- 7. Age of plant Maximum respiration occur in young leaves. Meristematic cells, germinating seeds etc. show high rate of respiration, because more energy is needed for cell division.

◆Pasteur's Effect

When the mode of respiration is changed from an aerobic to aerobic, there is a reduction in the consumption of respiratory substrate referred to as Pasteur's effect.

+Compensation Point

During the peak hours, the rate of photosynthesis is about 10 times higher than that of respiration. But at twilight (ie, twice a day ie, in the morning and evening) the rate of photosynthesis and respiration are equal. So that there is no exchange of gases between the plant and the environment, referred to as compensation point. So the ratio is 1:1.

If it is due to light intensity, then it is called light compensation point and if it is due to CO₂ concentration, then it is called CO₂ compensation point.

◆ **Ganong's Respirometer** - Apparatus which is used to measure the rate of respiration.