Plant Cell Bioprocessing
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Lecture – 05
Photosynthesis and Photorespiration

Let us study about photosynthesis. Photosynthesis is carried out by all green photosynthetic organisms which have chlorophyll in them. These organisms are called as autotrophs. They are capable of producing organic compounds and fixing the carbon dioxide using light energy.

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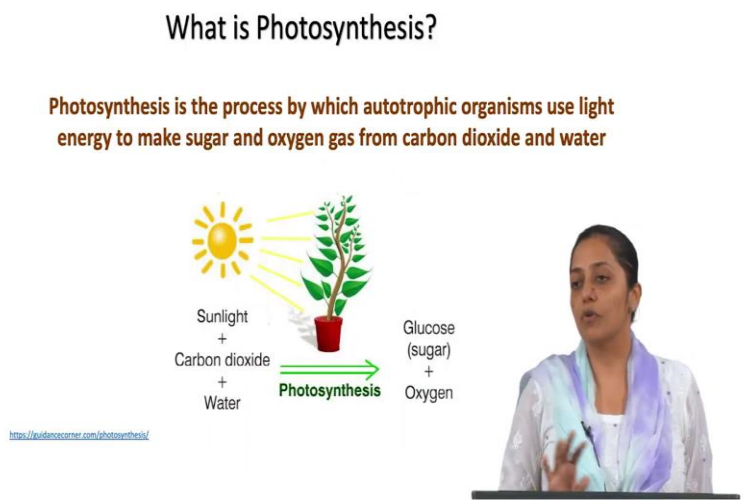

We all know in general what happens in photosynthesis, the light energy drives the production of sugars and oxygen from carbon dioxide and water.

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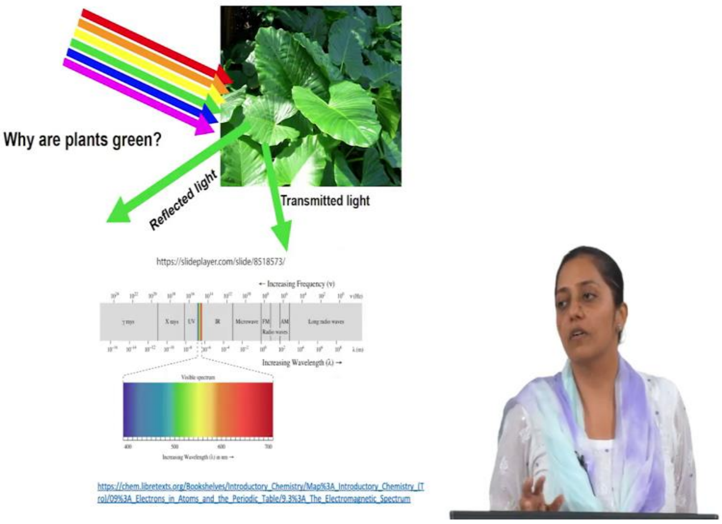

Why do plants look green? The rest of the light is absorbed and only green is transmitted and reflected.

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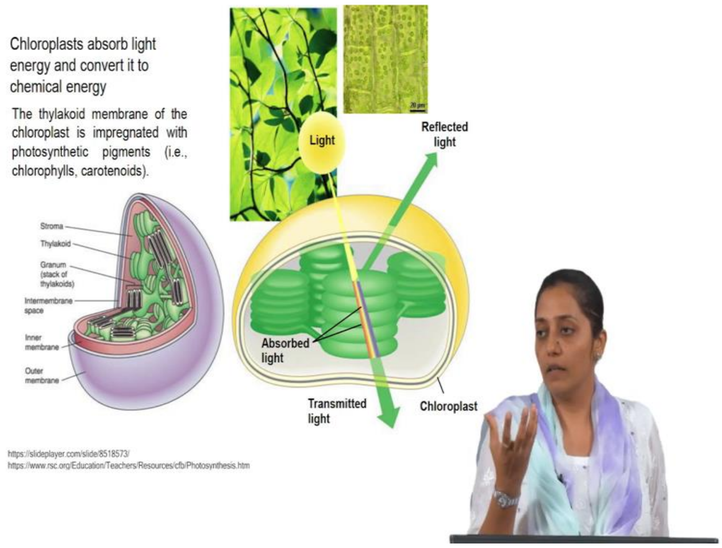

Photosynthesis takes place in chloroplast. The membranes of chloroplast are rich in a green coloured pigment called as chlorophyll. There are a number of granular stacks which are called as thylakoids. The light reactions happen in the membrane of these thylakoids and the carbon fixation happens at the stroma. The carbon dioxide diffuses into the stroma of the chloroplast. The light can easily pass through the chloroplast membrane.

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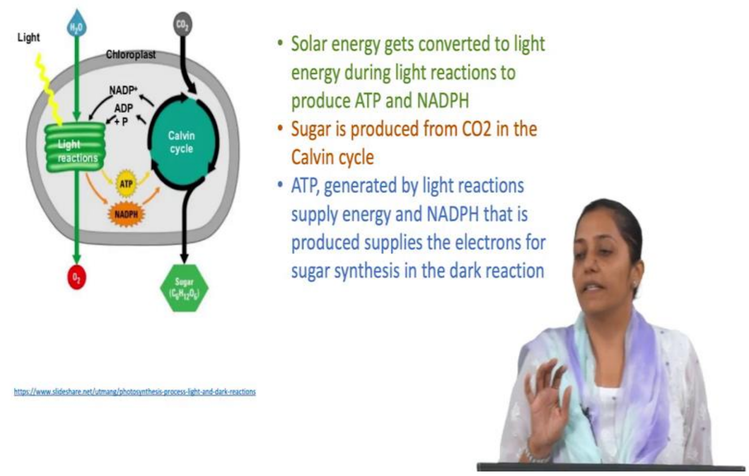

So, what happens? The entire process of photosynthesis is divided into two parts : light reaction and dark reaction. In light reaction, the light energy is captured by the photosystems which are called as reaction centres. These reaction centres have few 100 molecules of coloured pigments including chlorophyll. The reaction centres in these photosystems, photosystem 1 and photosystem 2 forms the acyclic phosphorylation cycle. Majorly in bacteria you will find cyclic phosphorylation. So, I am now going to talk about the light reactions of photosynthesis, where the water is broken down to produce oxygen and the photons are captured so as to drive ATP and NADPH synthesis.

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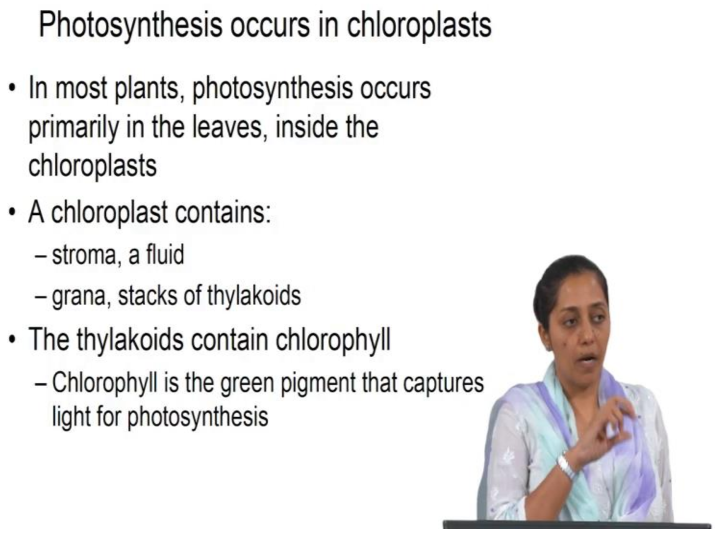

The stroma is the liquid part of the chloroplast and grana are the stacks of the thylakoid. The entire light reaction of capturing the photons and then passing on the electron excited electrons to the electron transport chain everything is present at the membrane of the thylakoid. Let us see what happens as soon as the light is incident on these reaction centres.

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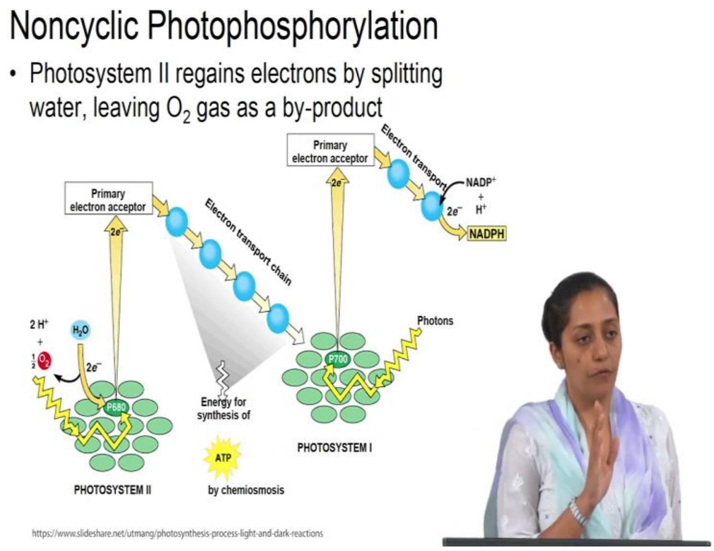

During evolution, among the two reaction centres, photosystem 1 came first, and then photosystem 2. But with adaptations to plants to make it more economical or more efficient, the two photosystems were brought in together. Therefore, photosystem 2 comes first and then the photosystem 1 in acyclic phosphorylation.

So, first is the photosystem 2 because it was discovered probably later. Light energy is incident on photosystem 2. There are number of pigment molecules and the energy which is absorbed by the pigment molecules keeps on getting transferred to neighbouring pigment molecules until it reaches the reaction centre. What is this reaction centre, it is a chlorophyll A molecule which can absorb light at a wavelength of 600 nanometres. So, it absorbs the energy and then this energy drives oxidation reduction reaction, breaking down the water molecule releasing, oxygen and the electrons.

Now, this excited electron is then captured by the neighbouring specific electron acceptor molecule. And then it subsequently keeps transferring the electrons in the electron transport chain which is present at the thylakoid membrane because of which the hydrogen ion gets released in the thylakoid lumen. So, now once the hydrogen ion concentration in the thylakoid lumen increases, an electrochemical gradient is generated.

So, this electrochemical gradient through chemiosmosis is then exploited by the ATPase which is present in the thylakoid membrane. The ATPase generates ATP by driving the hydrogen ions out in the stroma. The hydrogen ions which are let out in the stroma are combined with the cofactor NADP+ forming NADPH. So, this is how the light reactions give rise to ATP and the NADPH. Now, this ATP and the NADPH both are to be utilised for capturing CO_2 and for converting it into sugars.

Oxygen is an incidental by product because water is available and it is broken down to produce oxygen.

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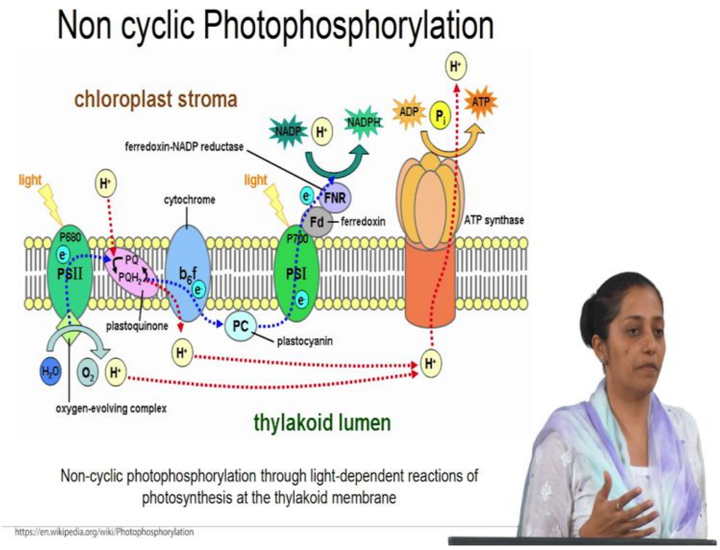

So, if you see the entire picture whatever I said would be very clear. These are similar to electron transport chain in mitochondria.

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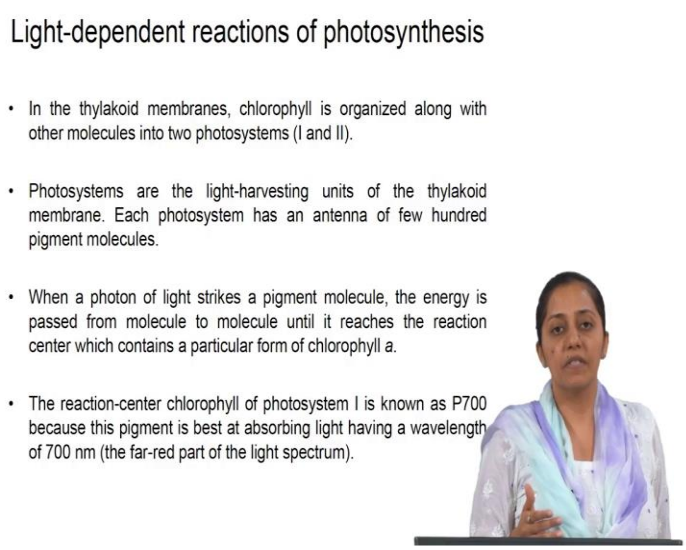

I will repeat the different steps in the thylakoid membrane. The chlorophyll is organised along with other molecules to from the two photosystems. Now, if you will observe that the photosystem 1 it is called as P700, photosystem 2 is P680 which means there is a difference in the maximum absorption wavelength. Now, it is a fact that both the reaction centres are identically same chlorophyll molecules A, but still there is a difference in the wavelength.

Why do you think? This difference in absorption properties happens because it is in conjugation with other proteins present in the reaction centre. So, because it combines with the other proteins in the two photosystems, there is a difference in the absorptive properties. Then the photosystems are light harvesting units. So, again to repeat there are two photosystems. When a photon of light strikes the photosystems, the electrons are released. These electrons being high energy states, are transferred to a nearing electron acceptor molecule which again transfers it to the electron transport chain present in the membrane, thereby releasing the hydrogen ions in the thylakoid lumen. This causes electrochemical gradient which is then exploited by ATPase to produce ATP, and the hydrogen ions sent back diffusing through the membrane into the stroma they are thenused to reduce the NADP+ to form NADPH.

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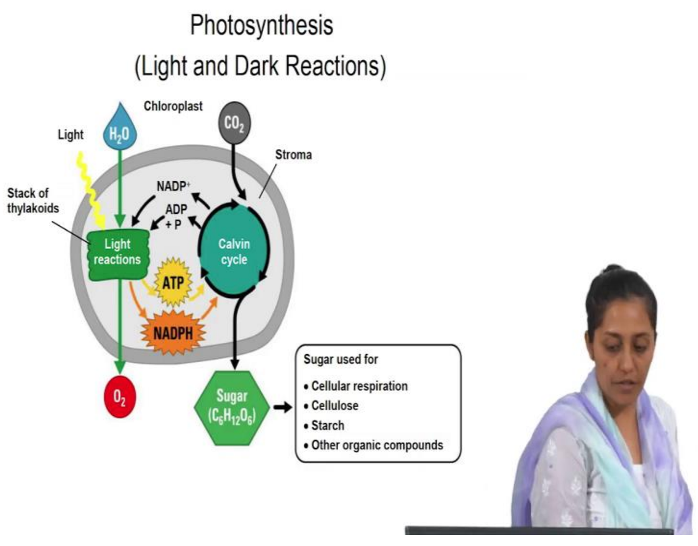

Let us talk about the dark reactions. So, this was the picture which I showed in the beginning. So, we have now spoken about the light reactions. Light reactions have given rise to ATP and NADPH. Now, dark reaction is called as Calvin cycle.

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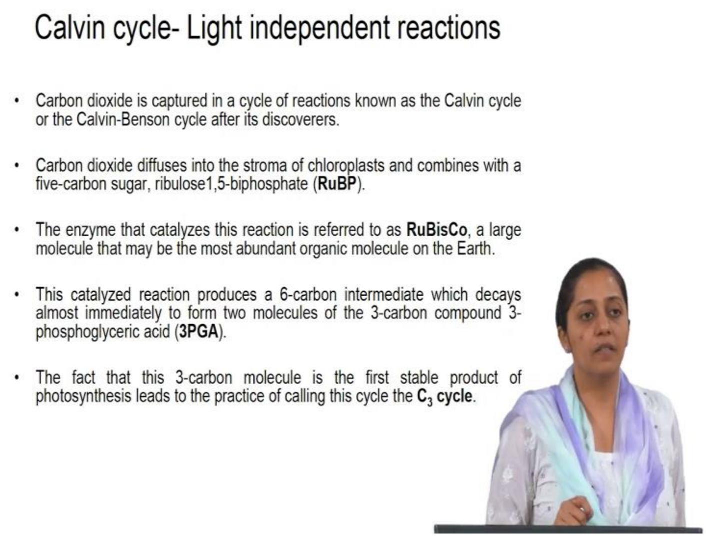

Now, why dark reactions? In this reaction the CO_2 is captured, this CO_2 is combined with ribulose 1, 5-bisphosphate. Now, in order to combine, you need an enzyme which is called as RuBisCo, it is the most abundant organic molecule present on the earth. So, this enzyme then combines the two to give you two stable and one unstable 6 carbon molecule which immediately breaks down to give you two stable molecules of phosphoglyceric acids. Now, because these are the first stable products of the Calvin cycle, this cycle is also called as C 3 cycle.

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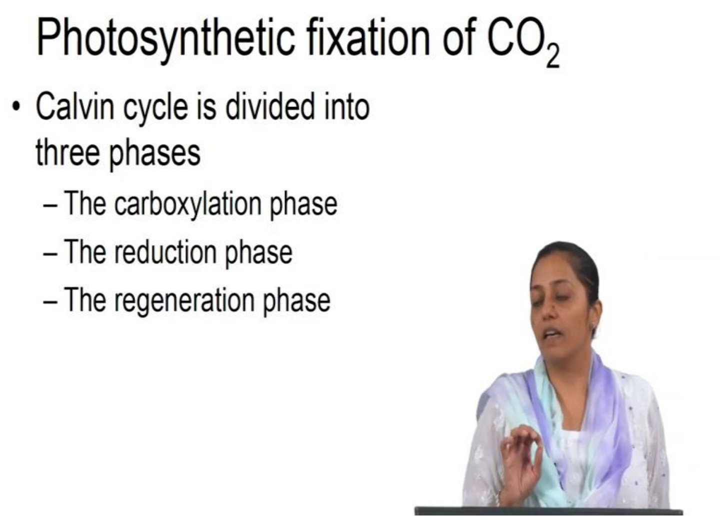

So, generally, in Calvin cycle, we have three portions, one is carboxylation, then reduction phase, and regeneration phase.

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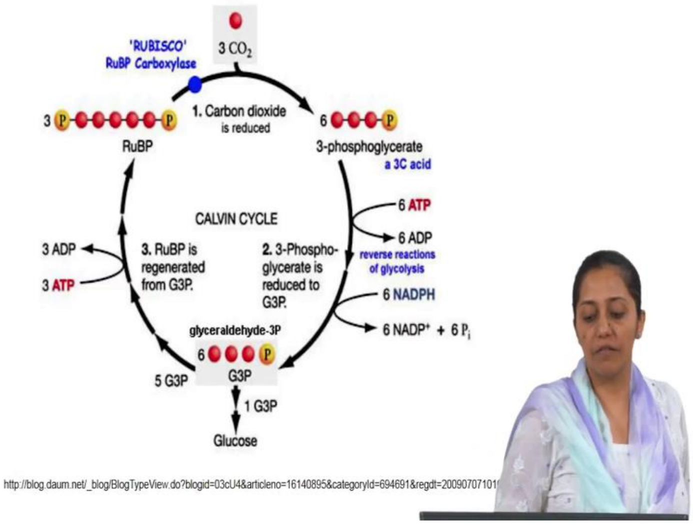

So, what happens in the carboxylation phase, the carbon dioxide is reduced by rubisco, which is ribulose bisphosphate carboxylase. So, it combines carbon dioxide with ribulose 1, 5-bisphosphate to give you two molecules of phosphoglyceric acid. Now, this phosphoglyceric acid is then reduced using ATP and NADPH and if you remember your gluconeogenesis which is reverse of glycolysis, some of these steps are similar to those reactions and such reactions takes place to convert your phosphoglyceric acid to 6 molecules of 3 phosphoglycerate.

Now, then one molecule is diverted to form your sugars and the remaining five is then again in the regeneration phase. Now, why it is called regeneration phase, because again the precursor molecular ribulose bisphosphate has to be regenerated. So, now, these five molecules are again regenerated to give you back ribulose bisphosphate which can then subsequently combine with CO_2 to give you PGA.

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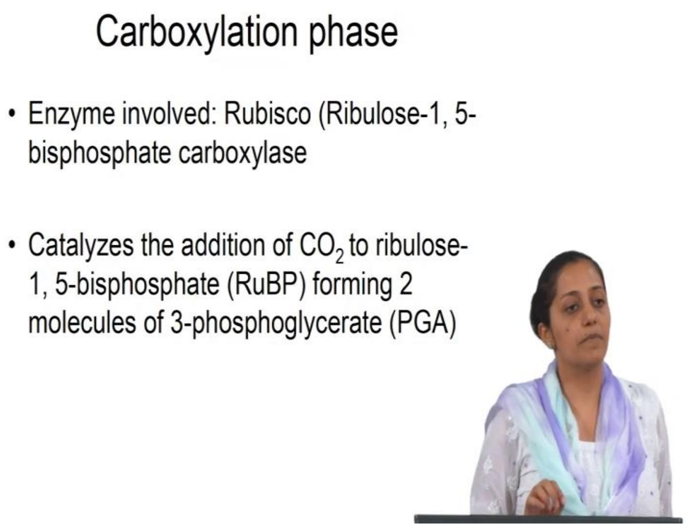

So, enzyme involved is Rubisco which is Rubisco 1, 5-bisphosphate carboxylase. Then, it catalyses the addition of CO_2 to ribulose 1, 5-bisphosphate as I said forming the first stable two molecules of phosphoglyceric acid.

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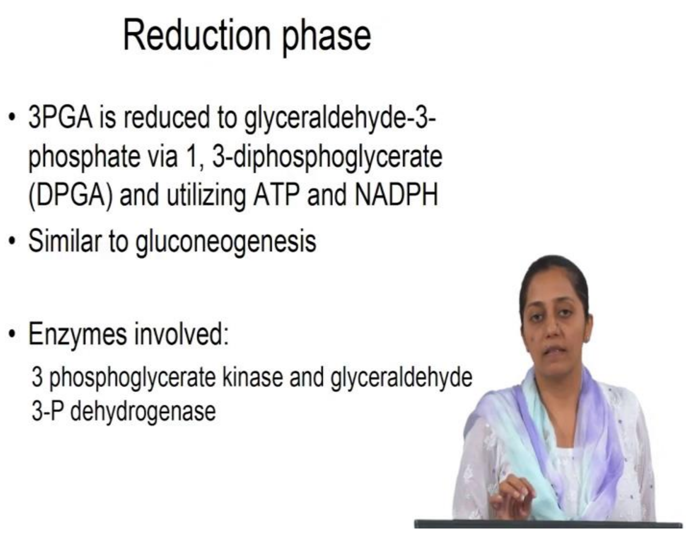

Then comes the reduction phase, as I discussed three phosphoglyceric acid is reduced to glyceraldehyde-3-phosphate. If you remember glyceraldehyde-3-phosphate is a part of glycolysis. So, it is then reduced to give you glyceraldehyde-3-phosphate which can then give you sugars. And there ATP and NADPH is needed to reduce PGA to glyceraldehyde-3-phosphate. Enzymes involved are 3-phosphoglycerate kinase which are the major enzymes and glyceraldehyde-3-phosphate dehydrogenase.

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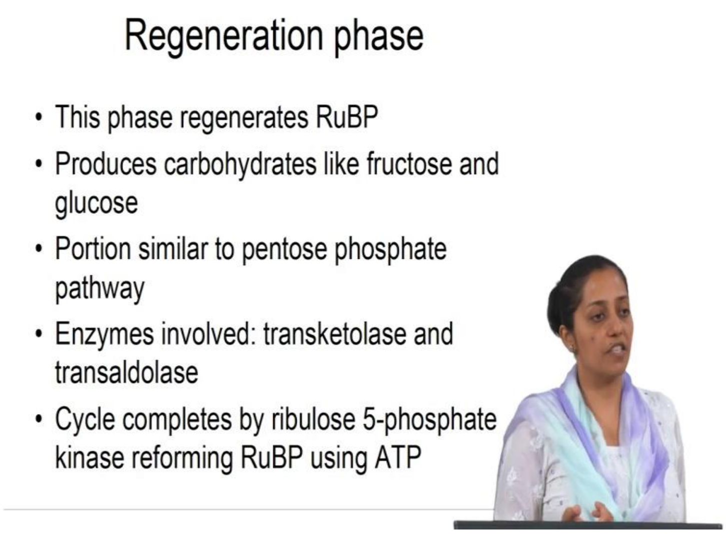

Now, comes the regeneration phase where the carbohydrates or fructose, and your glucose is produced, and how does it happens? The enzymes involved are transaldolase and transketolase. If you remember these are the enzymes which are involved in pentose phosphate pathway. So, again similar reactions of pentose phosphate pathways are involved. From glyceraldehyde-3-phosphate you get fructose diphosphate. If you remember your gluconeogenesis cycle of glycolysis.

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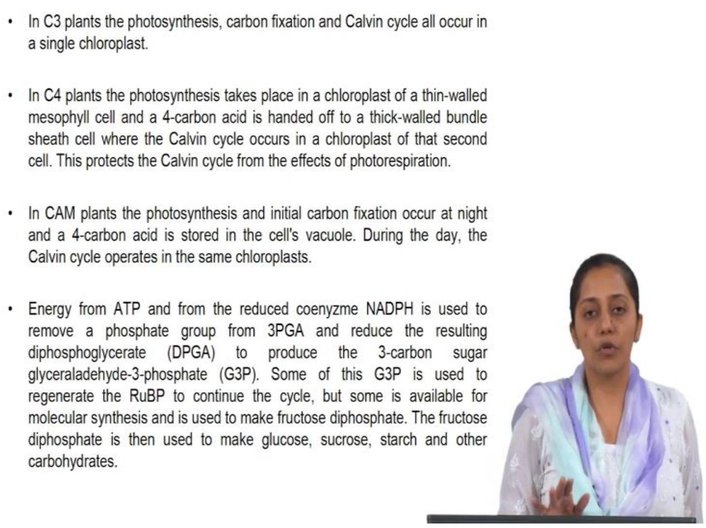

Now, coming back to adaptations. In C3 plants, generally, all the plants follow C3 metabolism. Now, in C3 plants, the light reactions and the dark reactions both are taking place in the same chloroplast, same cell. So, but what happens as I discussed earlier that in order to prevent excessive water loss, the stomata is closed. Now, once the stomata is closed, the CO_2, now is anyways getting utilised in carbon fixation.

So, then a time comes when the relative concentration of CO_2 to oxygen reduces. As soon as this happens Rubisco now starts getting more associated with oxygen. Now, what happens? Now, it starts acting as an oxygenase and it will combine your ribulose bisphosphate with oxygen. Now, this oxygen when it drives this reaction as oxygenase, it will give rise to a toxic molecule which is called as phosphoglycolate.

I will be talking about this process; now, once phosphoglycolate is formed, it is a highly toxic molecule. So, the cell drives certain reactions to convert this toxic molecule into a number of molecules like glycine, and then glycine gets converted to serine, and then serine thereby gives rise to many other molecules. Now, all these reactions takes place using organelles like peroxisome and mitochondria. Now, these reactions again happen at the expense of cell’s energy. So, ATP is involved and there is a loss of CO_2 when the glycine is getting converted to serine in mitochondria. So, effectively 30 percent of the carbon fixed is lost in salvaging this molecule.

So, we will see what do plants to in that case. So, there is one adaptation which is called as C4 metabolism. So, in C4 plants, the photosynthesis takes place in chloroplast and as I was mentioning that there are cells which surround the vascular bundle. These are called as bundle sheath cells. Now, why it is called C4? Because, the first stable compound formed is malate or aspartic acid. Now, this is light reaction, where this 4-carbon compound is then transported to bundle sheath cells where it again breaks down to release CO_2. Now, because it is now very close to vascular bundle and in the inner portions of the leaf of the tissue, it is set to be secured. Now, then once it releases CO_2 in the vicinity, the relative concentration of CO_2 to oxygen is then increased which then can cause Rubisco to again carry out the dark reactions. So, this is what C4 metabolism.Now, so there is a spatial change in the light and the dark reactions.

Now, then there is one other kind of metabolism which is called as CAM metabolism which is found in succulent plants. Now, why succulent plants, these leaves are plump with lot of water content, now there is a reason why do they become plump and with lot of water content because of the four C molecule which was malate.

Now, in these CAM metabolism, the stomata rather than opening in early hours of the morning and taking in CO_2 like C4 plants, these carry out the photosynthesis by capturing CO_2 to form a 4-carbon molecule at night. And in the morning, in the same cells, this molecule is broken down and photosynthesis or the dark reactions involving the fixation of the CO2 which has been released happens. So, it is a temporal shift between the light and the dark reactions.

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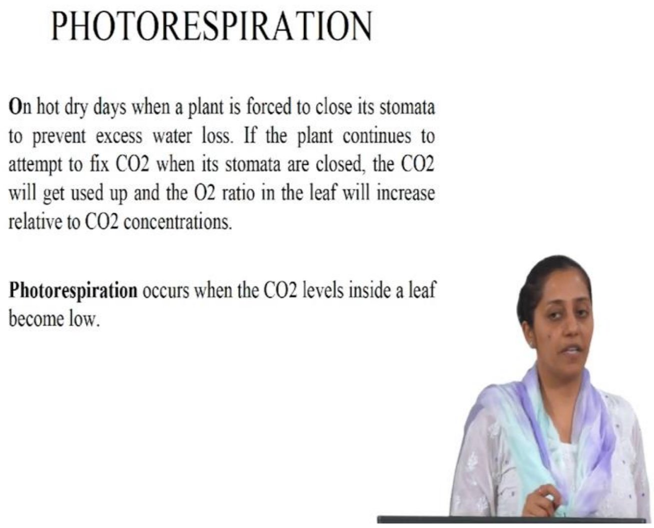

So, what happens in photorespiration, very hot days the stomata is closed. Once the stomata is closed, there is no gas exchange. The CO_2 is continuously getting utilised. Then again as I said relative concentration of CO_2 to O_2 is reduced. So, now, Rubisco is combining with O_2 to give you a toxic molecule phosphoglycolate. Now, this toxic molecules has to be broken down to prevent the cell damage. So, now, this whole process is called as photo respiration.

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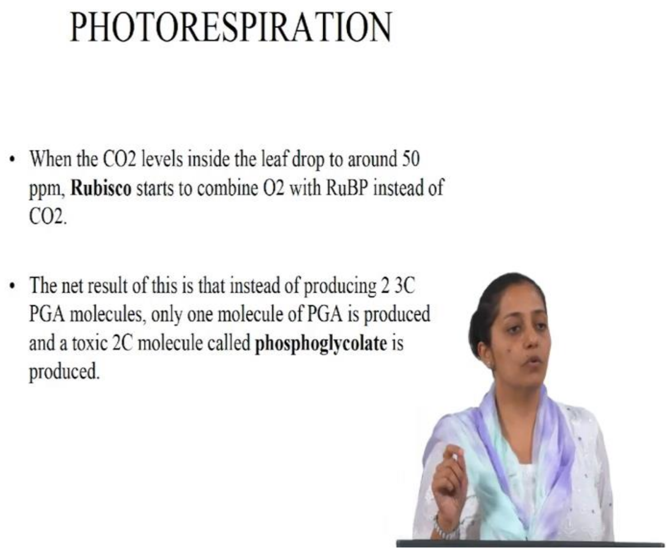

So, as I said see when the CO_2 levels drop in the leaf less than 50 ppm, then the Rubisco starts acting as an oxygenase.

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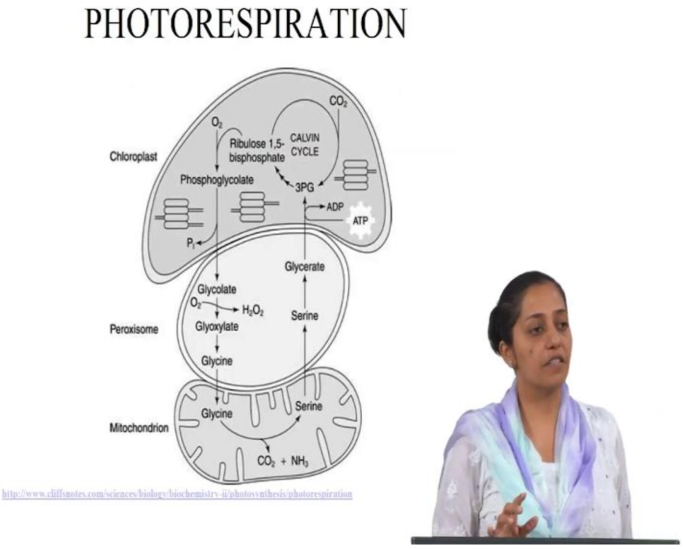

So, this are the reactions which take place in photorespiration. So, three organelles are involved chloroplast, peroxisomes and mitochondria. If you remember when I was discussing organelles, I asked you about the role of peroxisomes Peroxisomes play a crucial role in photorespiration. So, as I said this glycolate when formed is then sent to peroxisomes where the reaction to convert it into glycine happens. Now, this happens by utilising oxygen. So, in the peroxisome, the oxygen is then taken to convert your glycolate to glycine. Now, once the glycine is formed, it is then transported to mitochondria and then the glycine is converted to serine. Now, this reaction happens by taking in oxygen and by release of CO_2 so, therefore, respiration.

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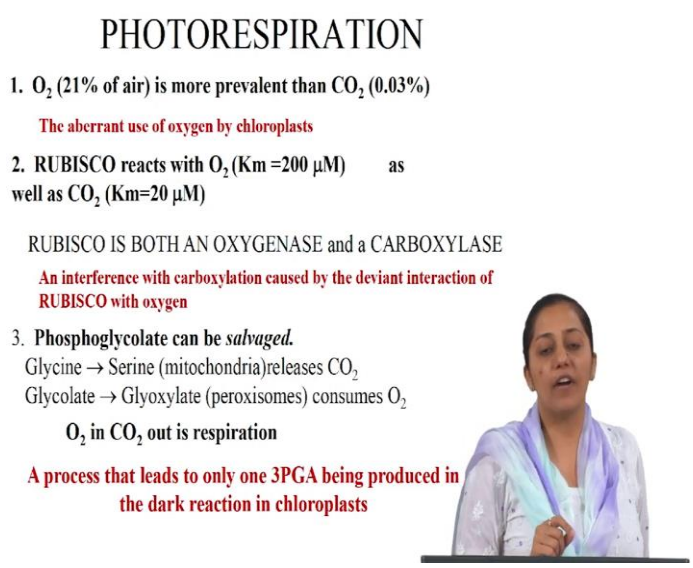

So, as I said the activity as oxygenase or carboxylase depends on their K_m value. The K_m although for Rubisco to combine with CO_2 is will be very low or very high. In general Rubisco combines with CO_2 so, if it can combine with oxygen the K_m value for CO_2 will be higher or lower? Lower, more affinity towards CO_2 than with oxygen.

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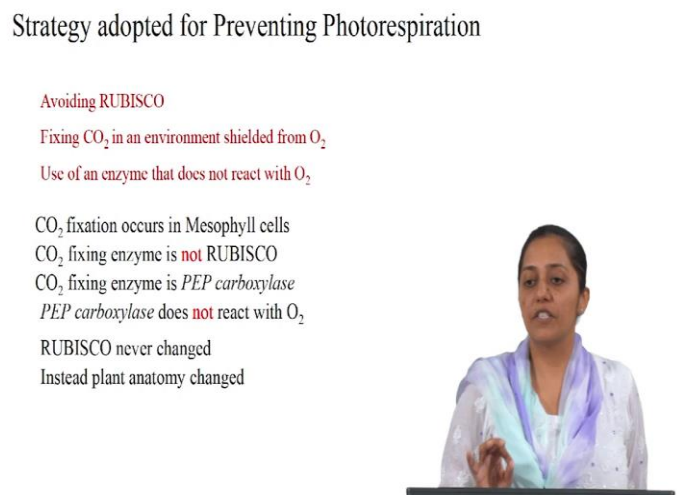

So, what in nature, the strategy could be applied to prevent this process photorespiration, either the plant should have avoided Rubisco or there should have been another enzyme to carry out photosynthesis or fixing CO_2 in an environment shielded from O_2, this could be another choice. The third choice could be use of an enzyme that does not react with O_2 which is the same. Do not use Rubisco or use another enzyme which does not react with O_2. So, this is what happened.

CO_2 fixation, as we know happens in the mesophyll cells, palisade cells, spongy cells with air gaps for gas exchange. So, now, CO_2 fixing enzyme is not Rubisco, the adaptation happened. Then CO_2 fixing enzyme was then changed from Rubisco to PEP carboxylase. Then Rubisco still remains the same, but CO_2 fixing enzyme became for the first light reactions it was changed to PEP carboxylase.

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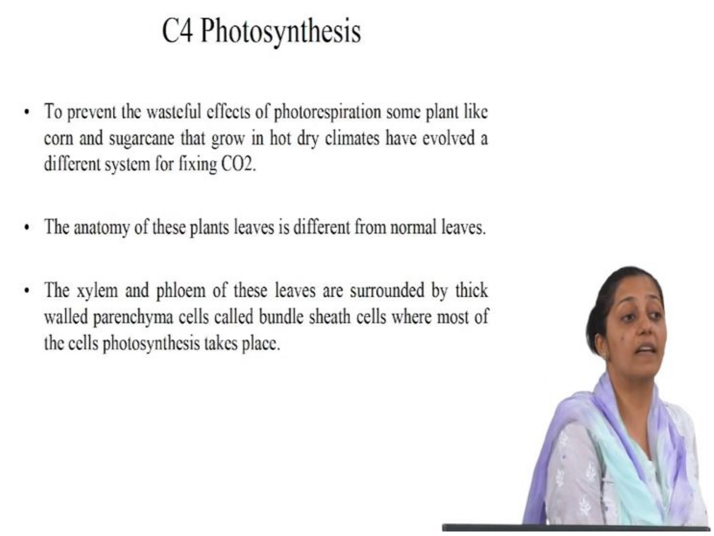

So, we will see how does the malate is formed. So, in C4 synthesis to prevent the wasteful effect of photorespirations, plants like your sugarcane and corn they adopt to this metabolism. Now, what happened, if you compare the anatomy between C3 and C4 plants, it is different. In C4 plants, you will see there are bundle sheath cells around the vascular bundle, which is xylem and phloem. While in your C3 plants, there are mesophyll cells, palisade cells, we saw columnar cells and then they were spongy cells. So, the chloroplast in C3 plants and all these cells can carry out light and dark both the reactions.

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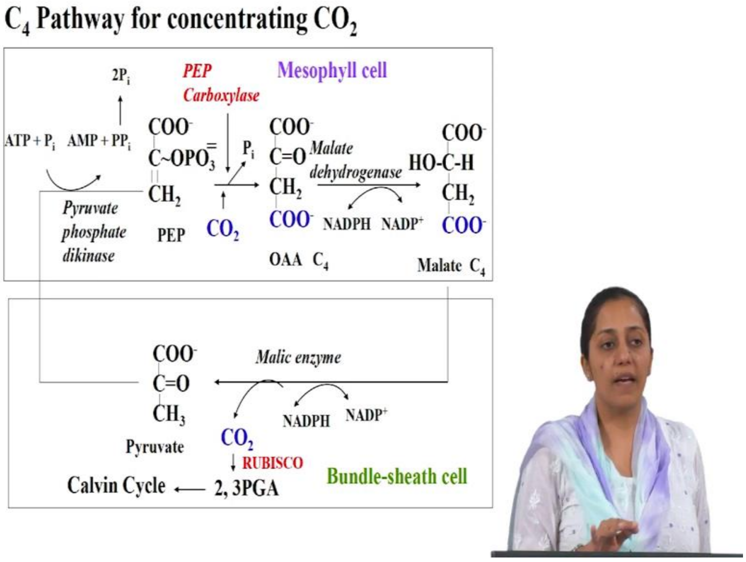

So, now, C4 pathway, is this where using the PEP carboxylase, PEP captures CO_2 to make the first four carbon molecule which is called as malate. This malate is then transported to bundle sheath cells in C4 plants where it is again broken down to release CO_2. Now, in this the Rubisco is protected, it is now not on the surface , so that it cannot be affected by the oxygen. So, here again then Rubisco combines with CO_2 in dark reactions to give sugar molecules.

Now, what happens in cam plants, I have already spoken about it, it is temporally different. So, this CO_2 fixation into malate happens early hours of the morning where CO_2 is taken in converted into malate, and then this during the day hours when the stomata is closed, this malate is broken down to give you fixing the CO_2, and then CO_2 combines with Rubisco to give sugars.

But then in CAM plants, at night, these reactions happen, the CO_2 is fixed to give you malate, malate is stored in vacuoles, and then in the morning in the malate is then broken down to give you CO_2. Now, this happens in succulent plants as I said. Now, why did I say succulent plants? They are more plump with lot of water, there is a large amount of malate present. So, lot of water is sucked in, so that is the reason why they become succulent in nature. And sometimes there is so much of pressure that the malate is forced to be broken down by such kind of plants.