**Introduction to Complex Biological Systems**
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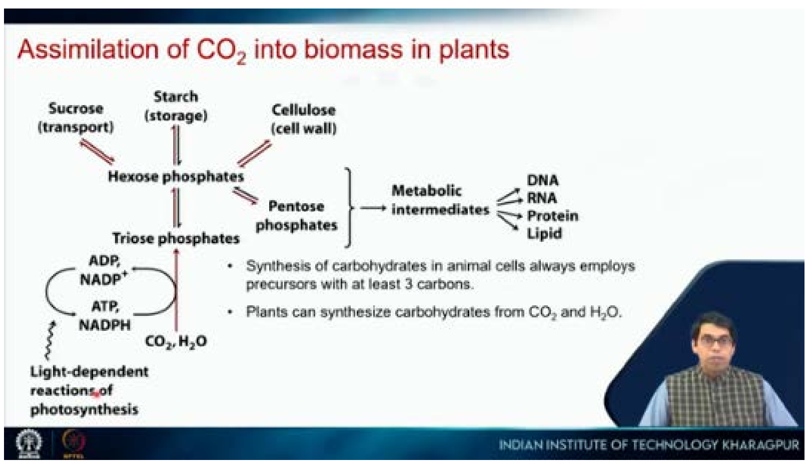
**Lecture 35**
**Photosynthetic carbohydrate synthesis**

Welcome to the course on introduction to complex biological systems. This is the last lecture of Week 7. Today, I am going to discuss photosynthetic carbohydrate synthesis. So, this is a summary slide that will show you what I am going to discuss today.

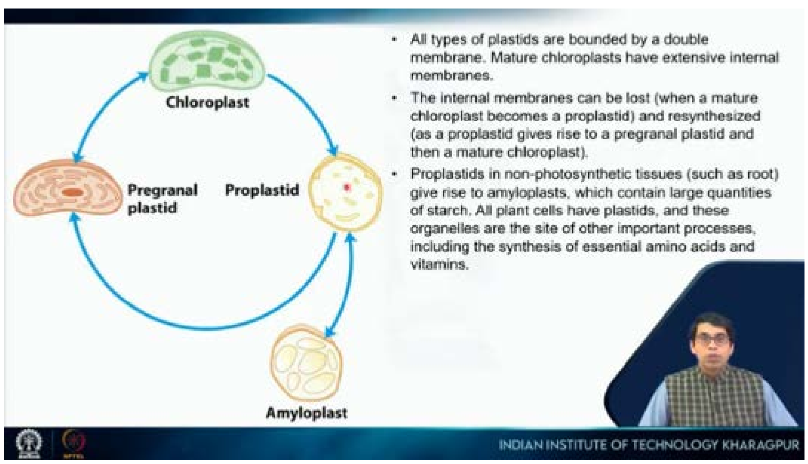
We have already seen the light-dependent reactions of photosynthesis. So, we have seen that light is used to generate these high-energy molecules. So, ATP from ADP and NADPH from $NADP^+$. So, these high-energy molecules are synthesized. That is something we saw in the last lecture.




Today, we are going to see that these high-energy molecules will be used to fix carbon dioxide. So, three molecules of carbon dioxide will be fixed to produce a three-carbon molecule, which is called a triose phosphate. So, this triose phosphate will then be the source for all these other molecules that are synthesized using various biosynthetic pathways. So, for example, the triose phosphate will be converted to a hexose phosphate. So, a six-molecule sugar.

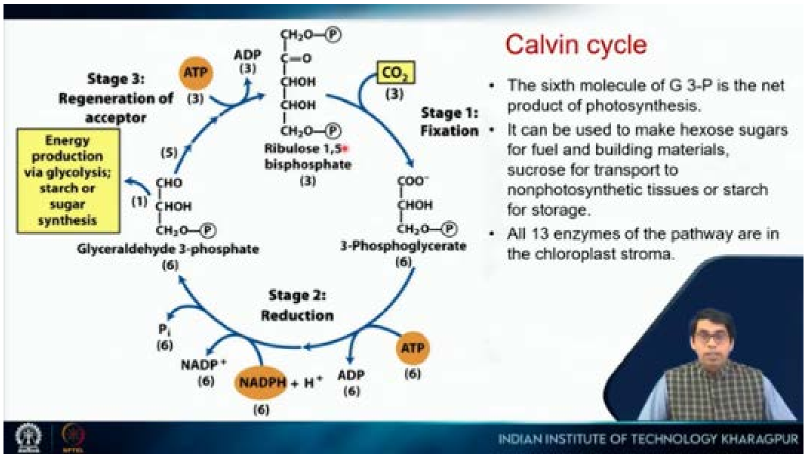
That hexose phosphate can be converted to fructose, which will then be converted to sucrose. This sucrose will be transported to different parts of the plant. The non-photosynthetic parts of the plant will use this sucrose as the sugar source. Hexose can also be stored as starch for storage. It can also be used to synthesize cell wall components or enter the pentose phosphate pathway that we have already seen. This pathway produces all these metabolic intermediates, which are used to synthesize DNA, RNA, proteins, and lipids. Now, in animal cells, the synthesis of carbohydrates always employs precursors with at least three carbons. So they start from here. These three-carbon molecules can come from the catabolism of glucose or any other carbohydrate source that we have consumed as food.

However, plants can synthesize these carbohydrates and all of these things directly from carbon dioxide. So plants can synthesize carbohydrates from carbon dioxide and water, and they use light as the energy source. In the last lecture, we saw chloroplasts as the site of action. Now I'm going to introduce another term, which is plastid. So it turns out that all types of plastids are bound by a double membrane.




We have seen that in the case of chloroplasts, and we have seen that in the case of mitochondria. So plastids are that, and mature chloroplasts have extensive internal membranes. So mature chloroplasts are also a type of plastid. However, mature chloroplasts have these extensive internal membranes that we saw in the last lecture, which are thylakoids. The internal membranes can be lost, as you see here, when a mature chloroplast becomes a proplastid, which is this and resynthesized, so they can be resynthesized, but it does not go like this. In this pathway, it goes to this pregranule plastid. So a protoplastid gives rise to a pregranule plastid and then a mature chloroplast. Protoplastids in non-photosynthetic tissues, such as roots, can give rise to amyloplasts, which are here and contain large quantities of starch. All plant cells have plastids, and these organelles are the site of important processes, such as the synthesis of essential amino acids and vitamins.

So all the essential amino acids and vitamins are synthesized in these amyloplasts. So now let us look at carbon fixation. Just like the citric acid cycle, this is another set of cyclic reactions. So here we are going to see carbon fixation and this cycle can be broadly divided into three stages.




Stage one, where the fixation of atmospheric carbon is done. Stage two, where reduction happens, and stage three, where this acceptor molecule, which is ribulose 1,5-biphosphate, is regenerated. So this cycle is called the Calvin cycle. So let us look at the number of carbons because we have to keep track of the number of carbons here. So it starts with three molecules of ribulose 1,5-biphosphate.

Now, this molecule has five carbons: 1, 2, 3, 4, and 5 and there are three molecules, so 5 times 3 will be 15. So, we start with 15 carbons. Three molecules of carbon dioxide come in, so that's another three carbons. So, 15 plus 3 is 18.

Now, these two together are converted into six molecules of 3-phosphoglycerate. Now, 3-phosphoglycerate is a 3-carbon molecule. So, 6 molecules of this 3-carbon molecule, so 6 times 3 is 18. So, we see that the number is conserved. 15 plus 3 was 18, and here we also have 18.

Now, these 6 molecules of 3-phosphoglycerate are reduced. So, this acid group is reduced to an aldehyde group. So, that is reduction, and that reduction is carried out by NADPH, which itself is oxidized to $NADP^+$. So, 3-phosphoglycerate becomes glyceraldehyde 3-phosphate. This is also a 3-carbon molecule, and we have 6 molecules of this.

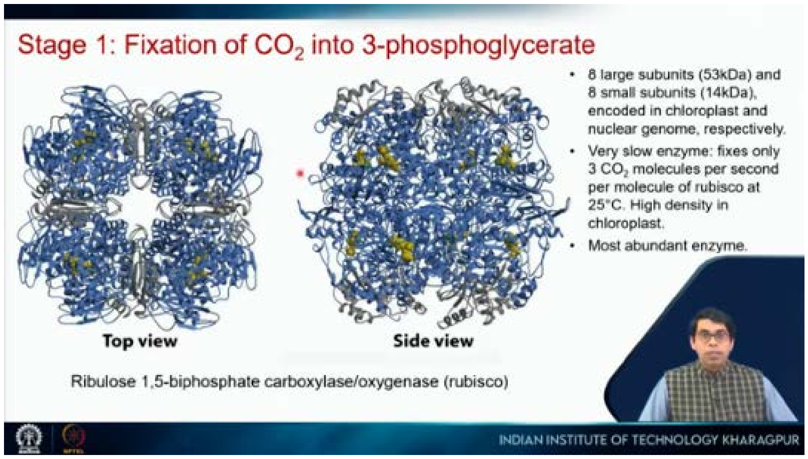
Now, at this stage, one molecule out of these six molecules goes out of this cycle, and this glyceraldehyde 3-phosphate can be used for energy production via glycolysis. We have seen that it is one of the intermediates in glycolysis, or it can be used for the production of sugar via gluconeogenesis or starch synthesis. The remaining five molecules will go back into this cycle. So, six molecules of glyceraldehyde 3-phosphate, that is 18 carbons, one goes out.

So, one molecule of glyceraldehyde 3-phosphate will have three carbons. So, you can see that these three carbons are typically balancing with these three carbons. So, I am not saying that these three carbons make this extra molecule, but you will see that these three carbons are balanced by these three carbons. So, you can sort of say that three carbon dioxide molecules now go out of this cycle as one molecule of glyceraldehyde 3-phosphate. Now, you are left with five.

So, five molecules which are each of three carbons, so five times three is fifteen, they are converted to three molecules of ribulose 1,5-bisphosphate. So, this is a five-carbon molecule and three molecules, so five times three is fifteen. So, this balance is also maintained.

So, essentially, the sixth molecule of glyceraldehyde 3-phosphate, the sixth molecule, is the net product of photosynthesis. So, these carbons are fixed, three carbons are fixed, and they go out of the cycle, and the remaining carbons are in the Calvin cycle. So, this can be used to make hexose sugar for fuel and building materials, such as sucrose for transport to non-photosynthetic tissues or starch for storage. So, this one glyceraldehyde 3-phosphate can be used for these purposes.

Now, all the 13 enzymes which are involved in this pathway are in the chloroplast stroma. So, this reaction or this cycle takes place in the stroma of the chloroplast. So, let us look at stage 1, which is the fixation of carbon dioxide into 3-phosphoglycerate. This reaction is carried out by this particular enzyme called ribulose 1,5-biphosphate, which is one of the substrates, and it is carboxylated. So, ribulose 1,5-biphosphate carboxylates.




However, we will see in a few slides that this enzyme also performs another reaction, which is called oxygenase. So, instead of fixing carbon dioxide, it will fix oxygen. So, that is why there is a slash here. So, ribulose 1,5-biphosphate. Either a carbon dioxide is added to it, or an oxygen molecule is added to it.

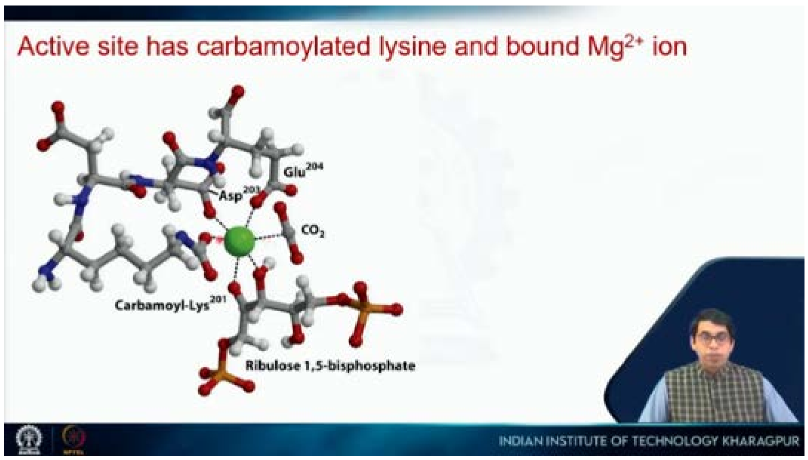
So that is why carboxylase/oxygenase and this long name of the enzyme is shortened as rubisco. So this enzyme is also called rubisco. This enzyme has 8 large identical subunits. Each subunit is 53 kilodaltons in size, and it has another 8 identical small subunits.

Each subunit is 14 kilodaltons in size. So, together it forms a very large protein complex. Interestingly, the large subunits are encoded in the DNA that is present in the chloroplast. So the chloroplast genome, whereas the small subunits are encoded in the nuclear genome.

So these two proteins are synthesized in different parts and then they come together and are assembled. This is one example of an enzyme that has a very slow turnover. So one molecule of rubisco fixes only three carbon dioxide molecules per second at 25 degrees centigrade. But this is a very important reaction. So it turns out that chloroplasts have a very high density of this molecule.

In fact, it turns out that among all the protein molecules present in a chloroplast, almost 50% of all protein molecules will be rubisco because you need this in very large quantities. This is similar to what we have seen in the case of hemoglobin. So the major part of the red blood cell is composed of hemoglobin. Similarly, here, almost 50% of all the proteins present in a chloroplast is rubisco. Since this is such an important reaction and it occurs in all different plants, rubisco makes up the most abundant enzyme on this planet.

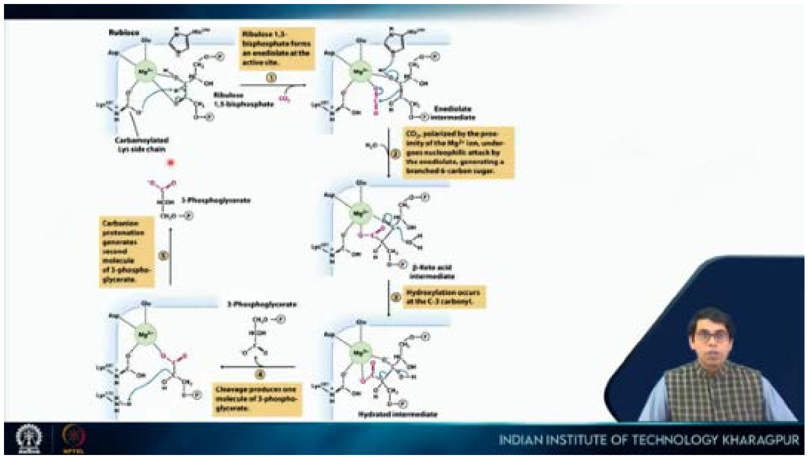
This is the active site of rubisco. So there are two important features that you should see. One is this lysine residue. So this is a lysine residue. You see the lysine side chain, and then it will end with an amine group.




So $NH_3^+$. However, this amine group is modified by a carbamoyl group. So this is carbamoylated lysine and there is a magnesium ion in the active site. So this magnesium ion is coordinated by this carbamoyl end of the lysine.

Then, these aspartic and glutamic acid side chains are there. The substrate is also there. This is the ribulose 1,5-bisphosphate and finally, carbon dioxide is there. So, this is the active site, and we will see how the reaction happens.

So, this is the carbamoylated lysine. So, that's the carbamoyl group. It is coordinating with the magnesium ion and this is your ribulose 1,5-bisphosphate. So, we can count the number of carbons: 1, 2, 3, 4, and 5. Now, this proton, so you see a ketone group here. The first reaction is the enol formation.




So you see that enol formation happens. So enol formation happens here. At this point, histidine 294 acts as a general base. So it abstracts this proton and triggers this reaction. Coordination of the carbon dioxide by the magnesium ion makes it a very good site for nucleophilic attack.

So this nucleophilic attack happens, and it is triggered by this. You can see the arrows are moving, so this proton is abstracted, the negative charge goes here, and these electrons attack this carbon. So this nucleophilic attack happens, and the carbon is now connected here. Now a water molecule comes in, and what it will do is cause the hydrolysis of this bond. So if we count the number of carbons at this point, 1, 2, 3 on one side, and 1, 2, 3 on the other side.

So it hydrolyzes this into two 3-carbon molecules. So this intermediate that is formed is called a beta-keto acid intermediate and now the hydroxylation occurs at this position, and this bond will break and this molecule is released. So this molecule is the first molecule of 3-phosphoglycerate that is released from rubisco.

So this is 3-phosphoglycerate. So this is carbon 1, carbon 2, and carbon 3. It is phosphorylated at that position. So it's a 3-phosphoglycerate. Now this rest part is still attached to the active site.

So a proton will be donated by this lysine to this carbonane and now it will be released as a second molecule of 3-phosphoglycerate. So, 5-carbon ribulose 1,5-biphosphate and 1 carbon dioxide, totaling 6 carbons, are converted to 2 molecules of 3-phosphoglycerate. Both have 3 carbons. So, 2, 3-carbon molecules are released.

That is the first step, which is the fixation of carbon dioxide, is complete. Now stage 2 is the reduction of this 3-phosphoglycerate to glyceraldehyde 3-phosphate. So you see this is the 3-phosphoglycerate. So 1, 2, 3, 3-phosphoglycerate. The first step is another phosphate group is added at this position.

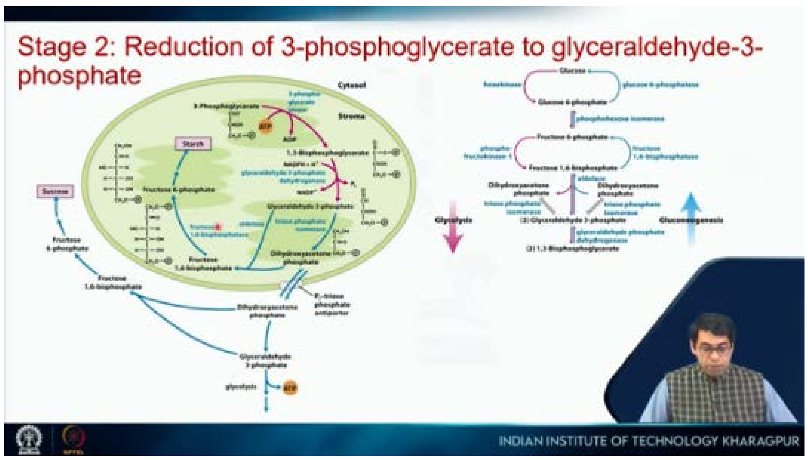
So it becomes an ester and then it is reduced by glyceraldehyde 3-phosphate dehydrogenase to glyceraldehyde 3-phosphate. So this acid group is reduced to an aldehyde group. So this becomes glyceraldehyde 3-phosphate. So again, 1, 2, 3, this is the third carbon so our second stage is complete. However, some more reactions happen, and it is important to show them here. Glyceraldehyde 3-phosphate can be converted to dihydroxyacetone phosphate. So again, it's a 3-carbon molecule. The only thing is that instead of an aldehyde, it becomes a ketone.

So this group is shifted. So it is done by an isomerase, which is triose phosphate isomerase. Now, one molecule of glyceraldehyde 3-phosphate and one molecule of dihydroxyacetone phosphate are condensed together by the enzyme aldolase to produce fructose 1,6-bisphosphate. Fructose 1,6-bisphosphate: one of the phosphate groups will be removed by fructose 1,6-bisphosphatase. So, the first phosphate group is removed.

So this phosphate group is removed, and it becomes fructose 6-phosphate, this molecule. Now, fructose 6-phosphate can be used to synthesize starch, which will be stored as the storage form of the sugar molecules. Another thing that can happen is this dihydroxyacetone phosphate can be transported out into the cytoplasm by this antiporter. It gets converted to glyceraldehyde 3-phosphate, and then this glyceraldehyde 3-phosphate, which is the same molecule here, can go into the glycolytic pathway. So it can produce ATP, and then it can produce further ATP molecules.

On the other hand, these two molecules can again be combined to form fructose 1,6-biphosphate like this, which gets converted to fructose 6-phosphate like this, and then it is converted to sucrose. Which will be transported to other non-photosynthetic parts of the plant, and there it will be kept as or used as an energy source. So all of these things can happen. However, we are interested in the formation of glyceraldehyde-3-phosphate. That will be one of the products of this stage.

Now, just for comparison, we have seen some of these reactions before. So, I am showing it to you here. This 1,3-biphosphoglycerate, this is 1,3-biphosphoglycerate. We have seen the formation of glyceraldehyde 3-phosphate from this. You see that is gluconeogenesis. So, glyceraldehyde phosphate dehydrogenase, the same enzyme is present here. Then triose phosphate isomerase converts it into dihydroxyacetone phosphate. Aldolase combines these two to form fructose 1,6-bisphosphate. Fructose 1,6-bisphosphatase converts it into fructose 6-phosphate.




So, these from here to here we have already seen in the case of gluconeogenesis. So, these are the same enzymes that you will see in different parts. So, we have glyceraldehyde 3-phosphate formation at this point. One molecule, so let us go back. So, six molecules of glyceraldehyde 3-phosphate are synthesized.

One molecule goes out. The remaining 5 will be used to regenerate 3 molecules of ribulose 1,5-bisphosphate. So, this is what we are going to see now. So, one molecule goes out like this or this. So, we will be left with 5 molecules of glyceraldehyde 3-phosphate and 5 molecules of glyceraldehyde 3-phosphate will be used to regenerate the ribulose 1,5-bisphosphate. That is stage 3. So three molecules of glyceraldehyde 3-phosphate are converted to dihydroxyacetone phosphate by triose phosphate isomerase. So let us say one molecule is converted to this, and one molecule remains as glyceraldehyde. Now these two are condensed by aldolase to form fructose 1,6-bisphosphate.

We saw this in the previous slide. So it is a 6-carbon molecule. It is converted to fructose 6-phosphate. We have also seen this in the last slide. So again, it is a 6-carbon molecule.

Now this 6-carbon molecule and another molecule of glyceraldehyde 3-phosphate. So remember we have 5 in our hand. This is 1, this is 2, this is the third one. These two combine. So this 6 plus 3, 9.

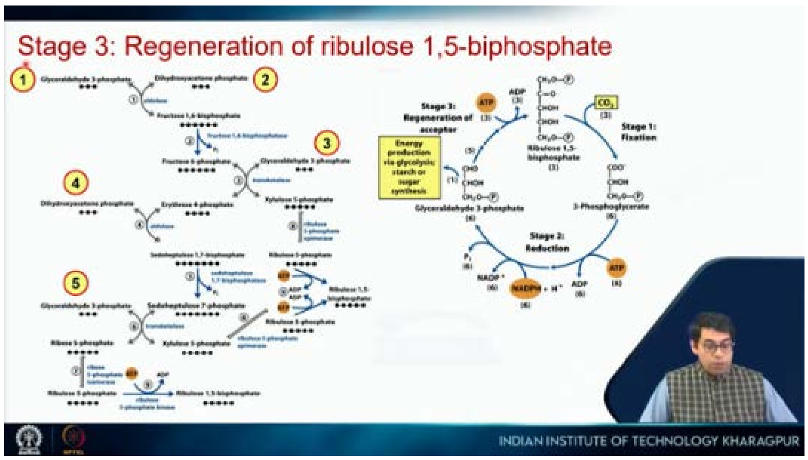
These two combine to form two sugar molecules. So six plus three, nine becomes four plus five, nine. So one molecule of erythrose 4-phosphate and one molecule of xylulose 5-phosphate. Now, see, we have a five-carbon molecule. We need a five-carbon molecule because ribulose is a five-carbon sugar.

So this is converted to ribulose 5-phosphate by ribulose 5-phosphate epimerase. So xylulose gets converted to ribulose, and then another phosphate group is added by this ATP to convert it to ribulose 1,5-bisphosphate. So we have synthesized one molecule of this. We need three. Now, the five-carbon molecule is used up here. This four-carbon molecule combines with another molecule of dihydroxyacetone phosphate, which comes from glyceraldehyde 3-phosphate. So four plus three is seven. These are condensed to form a seven-carbon molecule by the same enzyme, aldolase. You see, this is the same enzyme.So, it forms sedoheptulose-1,7-bisphosphate. Bisphosphate, one of the phosphate groups goes out, so it becomes sedoheptulose-7-phosphate. So, it is removed by this enzyme, sedoheptulose-1,7-bisphosphatase. So, we have a 7-carbon sugar and another molecule of glyceraldehyde 3-phosphate comes in. So, 7 plus 3 is 10. Now again, the transketolase comes in, the same enzyme that we saw here. So, it converts this 3 plus 7, 10-carbon into 2 5-carbon sugars. So, one is xylulose 5-phosphate, which is the same as this and again, it will be converted to ribulose 5-phosphate by the same enzyme, ribulose 5-phosphate epimerase and that is converted to ribulose 1,5-bisphosphate. So, from this path, we get the second molecule of ribulose 1,5-bisphosphate. Here, we have ribose 5-phosphate. This is converted to ribulose; this is ribose, this is ribulose. Ribulose 5-phosphate by the enzyme ribose 5-phosphate isomerase.

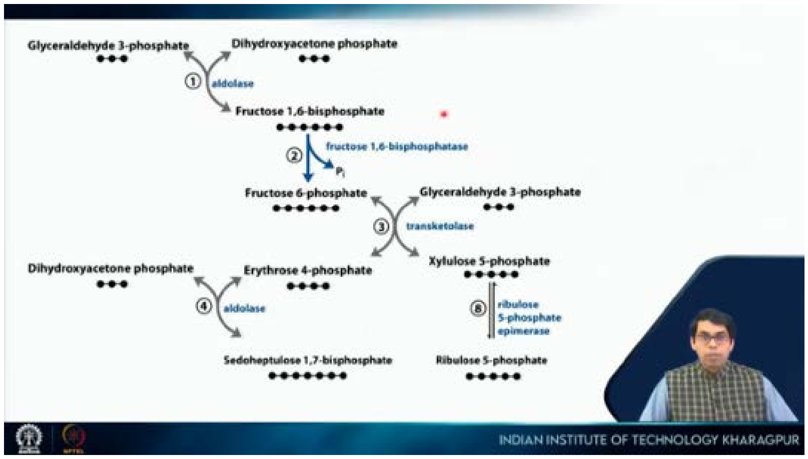
So now we have again ribulose 5-phosphate, just like this. So ribulose 5-phosphate kinase will use ATP to add another phosphate group to produce the third molecule of ribulose 1,5-

bisphosphate. So, two molecules of ribulose 1,5-bisphosphate came from here, and the third one is here. So, we used five molecules of glyceraldehyde 3-phosphate to produce three molecules of ribulose 1,5-bisphosphate. Where are the five molecules of glyceraldehyde 3-phosphate?

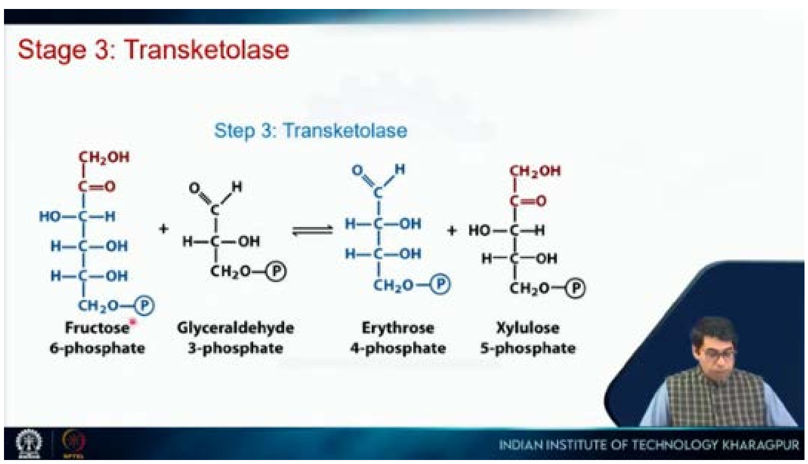
You can count them here. So, this is the first molecule. This is the second one because this comes from the same glyceraldehyde 3-phosphate. Third, fourth, and fifth. So these are the five molecules of glyceraldehyde 3-phosphate that produce one from here, two from here, and three from here.




Three molecules of ribulose 1,5-bisphosphate, which is this. So let us look at some of these steps, these steps a little more carefully, so that we can look at the actual structures of these molecules. So if we look at this step 3, fructose 6-phosphate and glyceraldehyde 3-phosphate. So this is fructose 6-phosphate, 1, 2, 3, 4, 5, 6 carbons, and this is glyceraldehyde 3-phosphate. What transketolase does is it will take these two carbons and put them here.

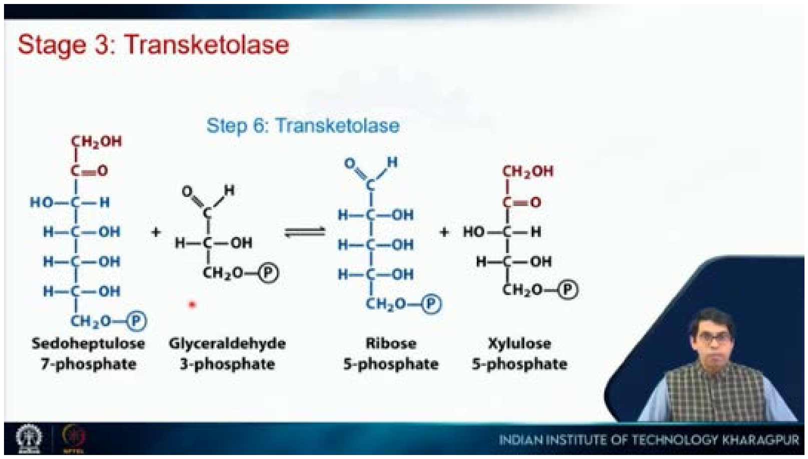



So now we have a 4-carbon sugar, erythrose, and a 5-carbon sugar, xylulose. So erythrose and xylulose are formed and then it will go through two different paths. Erythrose combines with this to form aldolase, which is a seven-carbon molecule and then that seven-carbon molecule combines with another three-carbon to form two five-carbon molecules.

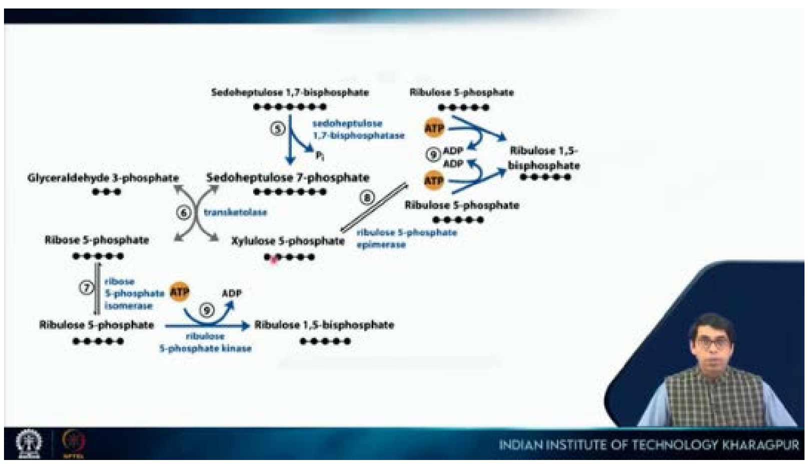



So this is again catalyzed by transketolase. So let us look at that. So this is the 7-carbon molecule. 1, 2, 3, 4, 5, 6, 7 and 3 carbons from the glyceraldehyde 3-phosphate. 1, 2, 3. So 7 plus 3 is 10.

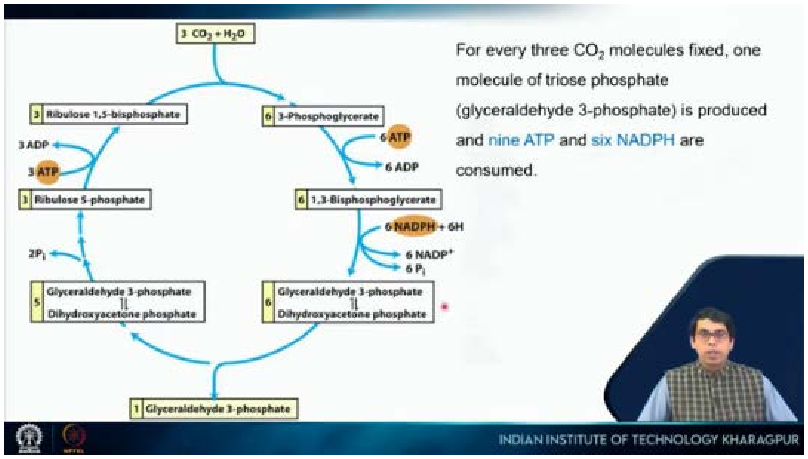
Now it forms 2 five-carbon molecules. Ribose. 1, 2, 3, 4, 5 and xylulose and then these two go into these two pathways.




So, xylulose will get converted to ribulose and finally ribulose 1,5-biphosphate. Ribose also gets converted to ribulose and finally ribulose 1,5-biphosphate. So, let us look at the energy usage for the production of glyceraldehyde 3-phosphate from three carbon dioxide molecules. So, remember in the Calvin cycle, three carbon dioxide molecules produce one glyceraldehyde 3-phosphate.




So, for every three carbon dioxide molecules fixed, one molecule of glyceraldehyde 3-phosphate is produced and to do that, 6 plus 3, 9 molecules of ATP and 6 molecules of NADPH are used. So, very high energy is used to produce this one molecule of glyceraldehyde 3-phosphate. Now, if you remember from the previous lectures this week, we saw that NADPH and NADH are similar.

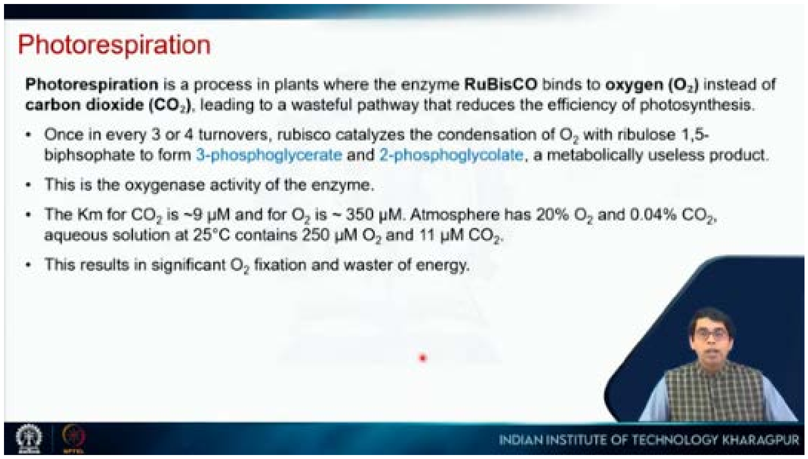



The only difference is a phosphate group on the sugar molecule. So, if I assume NADPH as equivalent to NADH and we also see that NADH is equivalent to 2.5 molecules of ATP. So, 6 molecules of NADPH will be equivalent to 6 times 2.5, that is 15 molecules of ATP. So, 15 here and 9 here, that is 24 molecules of ATP, that is the equivalence of ATP, are used to produce 1 molecule of glyceraldehyde 3-phosphate from 3 carbon dioxide molecules.

So, 24 equivalents of ATP are used. If we think of the production of glucose from this, then two molecules of glyceraldehyde 3-phosphate will be needed to produce one glucose molecule. So, in essence, 24 times 2, 48 molecules of ATP will be consumed to produce one glucose molecule from six carbon dioxide molecules. So we are using up 48 equivalents of ATP. Now, if you remember the complete oxidation of glucose via glycolysis, the citric acid cycle, and oxidative phosphorylation, it produces 32 molecules of ATP.

So here we are using, consuming 48 molecules of ATP to produce glucose and when we oxidize glucose back to carbon dioxide and water, we produce 32 molecules of ATP. So 32 out of 48 is two-thirds or 66%. So we get back 66% of the energy that is used to produce glucose, which is not bad.

That is very good efficiency that we can actually get back 66% of the energy that has been used to produce this high-energy molecule like glucose and ultimately, all of this energy is coming from the sun. So solar energy is the source. Now let us look at this other part that rubisco does. So it fixes carbon dioxide.




That is what we have seen. But it can also fix oxygen to this molecule, ribulose 1,5-biphosphate and that process results in something which is called photorespiration. So photorespiration is a process in plants where the enzyme rubisco binds to oxygen instead of carbon dioxide, leading to a wasteful pathway that reduces the efficiency of photosynthesis. So it turns out that in every 3 or 4 turnovers, rubisco catalyzes the condensation of oxygen with ribulose 1,5-biphosphate.

So instead of carbon dioxide, it uses oxygen to form 3-phosphoglycerate and 2-phosphoglycolate. So remember that when ribulose 1,5-biphosphate, which has 5 carbons, is fixed with carbon dioxide, totally 6, it will produce 2 molecules of 3-phosphoglycerate. But instead of carbon dioxide, if it is oxygen, we do not have this extra carbon. We have only five carbons from the ribulose 1,5-biphosphate. So three of them produce 3-phosphoglycerate, and the remaining two produce 2-phosphoglycolate, which is a two-carbon molecule and this two-carbon molecule is a metabolically useless product. So it has to be reconverted back to 3-phosphoglycerate and that uses a lot of energy. So this is a wasteful expenditure of the energy that was tapped by these light reactions during photosynthesis.

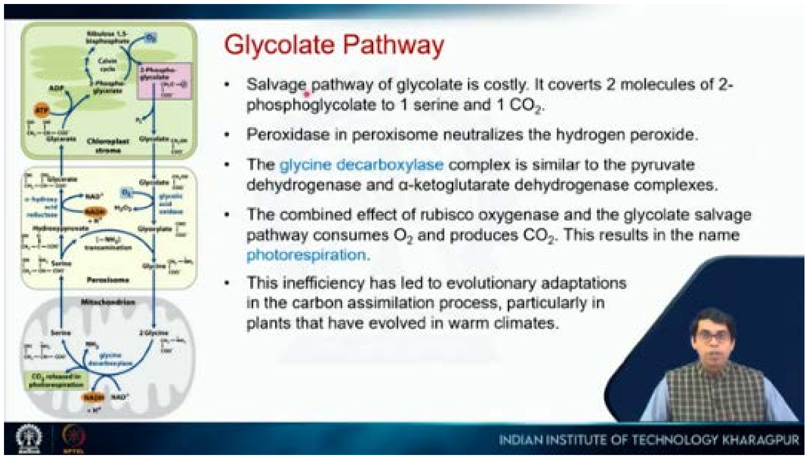
So this fixing of oxygen is the oxygenase activity of the enzyme. That is why this enzyme is called a carboxylase/oxygenase. The $K_M$ or the Michaelis constant for carbon dioxide of rubisco is around 9 micromolar, whereas that of oxygen is 350 micromolar. So it binds carbon dioxide more tightly than oxygen.

However, if you think about the substrate concentration, we have more oxygen than carbon dioxide. So the atmosphere has 20% oxygen and around 0.4% carbon dioxide and if you think about the dissolved gas, then the concentration of oxygen is 250 micromolar, whereas that of carbon dioxide is 11 micromolar. So if you have a high substrate concentration, then of course the saturation of the enzyme will also be high.

So since we have a higher concentration of oxygen, even though its Michaelis constant is less, we will still get a higher saturation, almost equivalent to that of carbon dioxide. So that is why in every 3 or 4 turnovers, one is an oxygenase reaction that is catalyzed by rubisco and this results in significant oxygen fixation and a waste of energy. So this is the reaction, the ribulose 1,5-biphosphate.


So this is the enol in diol form that we saw. Now oxygen binds here instead of carbon dioxide and then it produces these two molecules. So this is the good product, 3-phosphoglycerate, which can be used. But this is the useless metabolite, which is 2-phosphoglycolate.

So this has to be somehow salvaged and the salvage pathway is called the glycolate pathway. So this salvage pathway of glycolate is costly. It converts two molecules of 2-phosphoglycolate to one molecule of serine and one molecule of carbon dioxide. So two molecules of phosphoglycolate means 4 carbons, serine has 3 carbons, and the remaining carbon goes out as carbon dioxide, which will again be captured by rubisco to do its fixation. Now, the hydrogen peroxide that is produced, so we will see hydrogen peroxide is produced here. It is neutralized by peroxidase enzymes in the peroxisome. So, let us look at the reaction. Let us say rubisco takes up oxygen instead of carbon dioxide.




It fixes it with ribulose 1,5-bisphosphate to produce one molecule of 2-phosphoglycolate, which is not a useful molecule. So, this 2-phosphoglycolate has its phosphate group removed by phosphatase to produce glycolate. This is $CH_2H$ and $CO^-$. So, this molecule now moves out from the chloroplast into the peroxisome.

Here, the glycolate is oxidized to glyoxylate using oxygen. So, glycolic acid oxidase is the enzyme that produces this glyoxylate. So, the alcohol group is oxidized to an aldehyde group. Now, a transamination reaction occurs, which adds this $NH_2$ group here and converts it into glycine.

We have already seen this amino acid. So, $CH_2$ $NH_3$, and $CO^-$. So, this is glycine. This glycine now passes out from the peroxisome and goes into the mitochondria. So, you see that it is passing through all these different organelles.

Two molecules of glycine are taken, and glycine decarboxylase is involved. This is an enzyme complex. So there are three enzymes, and this enzyme complex takes up two molecules of glycine, removes ammonia, releases one carbon dioxide, and converts it into serine. So you see there are two carbons. So two glycine molecules mean four carbons.

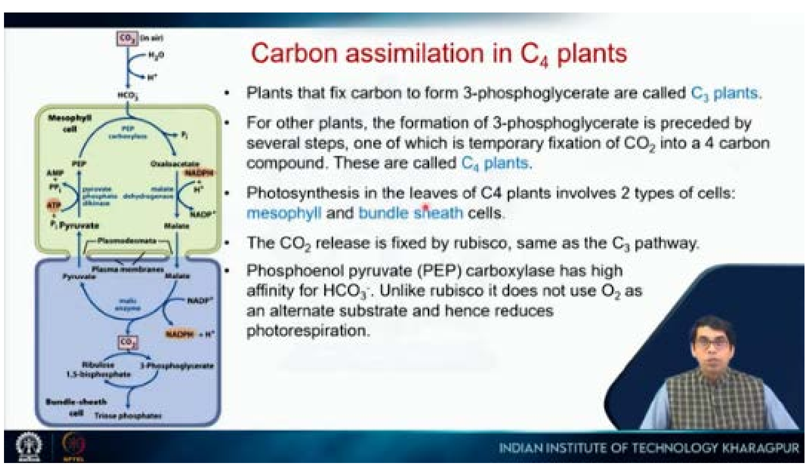
Those four carbons are converted into serine. So you have one, two, three carbons, and the remaining carbon goes out as carbon dioxide. So that is why this is called photorespiration. What happens in respiration? We consume oxygen and release carbon dioxide.

So here, oxygen is consumed, and carbon dioxide is released. So this is equivalent to respiration and to distinguish it from normal respiration or cellular respiration that happens in mitochondria, this is called photorespiration. Now the serine molecule is passed back to the peroxisome, where the amine group is taken off.

So deamination happens. So this amine goes from here to the glycine, and it forms hydroxypyruvate and this hydroxypyruvate is now reduced to glycerate by this enzyme alpha hydroxy acid reductase. So it uses NADH as the reducing agent, which gets converted to $NAD^+$. And this glycerate goes into the chloroplast, where it is phosphorylated to produce 3-phosphoglycerate. So this 2-phosphoglycolate finally produces this 3-phosphoglycerate. Now, two molecules of these are taken to produce one molecule of 3-phosphoglycerate. Three carbons are here, and one carbon is released as carbon dioxide.

So the combined effect of rubisco oxygenase and the glycolate salvage pathway consumes oxygen, which is here, and produces carbon dioxide, which is here. This results in the name photorespiration. This inefficiency has led to evolutionary adaptations in the carbon assimilation process, particularly in plants that have evolved in warm climates. Because in warm climates, again, the binding of carbon dioxide becomes weaker.

So photorespiration becomes even more dominant. So let us look at the other pathways. So one is carbon assimilation in $C_4$ plants. So plants that fix carbon to form 3-phosphoglycerate, as we have seen till now, these plants are called $C_3$ plants. They are producing a three-carbon molecule.



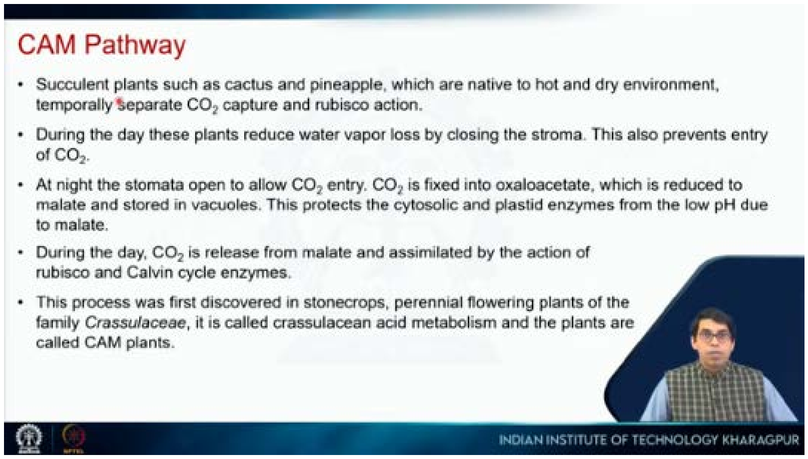
There are other plants where the formation of these three phosphoglycerides is preceded by several steps, one of which is the temporary fixation of carbon dioxide into a four-carbon compound. So they produce a four-carbon compound, and these types of plants are called $C_4$ plants. Photosynthesis in the leaves of $C_4$ plants involves two types of cells. Mesophyll, which is shown here in green, so this is mesophyll, and the other one is bundle sheath cells, which is shown here in blue. So here what happens is phosphoenolpyruvate. So this phosphoenolpyruvate in the mesophyll cells combines with bicarbonate ions.

So carbon dioxide is dissolved in water to produce these bicarbonate ions and phosphoenolpyruvate carboxylase has a very high affinity for these bicarbonate ions. So it does not have any affinity or much affinity for oxygen. So here a filter works. So phosphoenolpyruvate is converted into oxaloacetate by this enzyme, phosphoenolpyruvate. Now oxaloacetate is converted to malate, which passes on into the bundle sheath cells. Here it is oxidized to pyruvate and one molecule of carbon dioxide goes out. This pyruvate goes back into the mesophyll cells.

Phosphorylation of this happens by this enzyme, pyruvate phosphate dikinase and it produces this phosphoenolpyruvate. So it regenerates this phosphoenolpyruvate. So the net effect is that carbon dioxide is dissolved as bicarbonate, which goes into this cycle and is released here and this carbon dioxide is taken up by rubisco to produce 3-phosphoglycerate.

Now this whole pathway eliminates oxygen from this, so rubisco is not coming in contact with oxygen and hence works at its maximal efficiency. So this reduces photorespiration in $C_4$ plants. There is another pathway which is called the CAM pathway. So succulent plants such as cactus and pineapple, which are native to hot and dry environments, temporally separate carbon dioxide capture and rubisco action.

So the carbon dioxide capture that happens and its fixation by rubisco are separated in time. So how do they do that? So during the day when it is very hot, these plants reduce water vapor loss by closing the stroma. So if you close the stroma, then of course, no exchange of gaseous carbon dioxide or oxygen is happening. So this prevents the entry of carbon dioxide also.

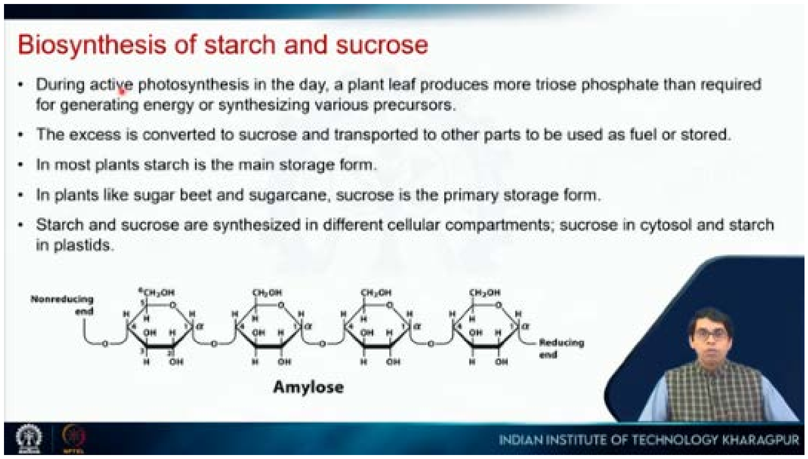



No water is released, and no carbon dioxide is taken up either. However, at night when the temperature is low and hence the loss of water vapor will be less, the stroma opens, allowing the entry of carbon dioxide. Carbon dioxide is fixed into oxaloacetate, which is reduced to malate as we saw in the previous slide and then this malate is stored in vacuoles. So the malate is produced, and it is not immediately used to release carbon dioxide.

So this malate is stored in special organelles called vacuoles. Why store it in vacuoles? Because malate, which is malic acid, will release protons and reduce the pH and this reduction of pH can damage other enzymes present in the cytosol or the plastid. So to protect those enzymes, this malate is stored in vacuoles.

During the day, carbon dioxide is released from the malate as we saw here. So from the malate, carbon dioxide is released. This is happening during the day when the stroma is closed, so no carbon dioxide is entering, and no oxygen is entering either. So this is released from the malate, assimilated by the action of rubisco and Calvin cycle enzymes. So by this temporal separation of carbon dioxide capture into malate and release of carbon dioxide to rubisco, When the stroma is closed, so no oxygen is coming in, it reduces photorespiration. This process was first discovered in stonecrops, such as the family of flowering plants called crassulaceae.

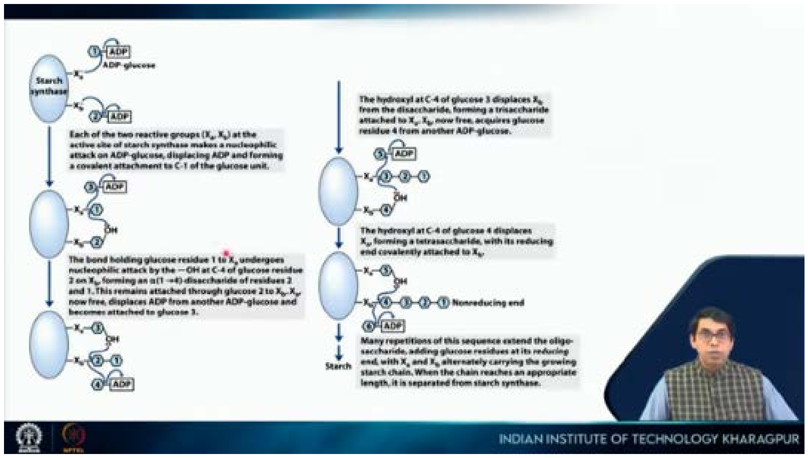
So that is why, based on this, these are called CAM plants, and this pathway is called the CAM pathway. So the last part that we will see is the biosynthesis of starch. So during active photosynthesis in the day, a plant leaf produces more triose phosphate, which is glyceraldehyde 3-phosphate, than it requires for generating energy or producing other biosynthetic materials. So the excess molecules are converted to sucrose and transported to other parts of the plant to be stored or used as fuel and another part is stored as starch.




So starch is the main storage form. So in plants like sugar beet and sugarcane, sucrose is the primary storage form. So starch and sucrose are synthesized in various different cellular compartments. Sucrose is produced mostly in the cytosol, and starch is produced in plastids. So this is something that we have seen earlier.

So I can go back to that slide and show you. So sucrose is produced in the cytosol, and starch is produced in the plastid. So this is an example of a starch molecule. So this is amylose. So it's a linear chain of sugar molecules and typically, when you study sugar chemistry, you will see that this end is referred to as the reducing end of the sugar and this end is called the non-reducing end. I'm just introducing these two terms because this will be useful in the next slide where we see the synthesis of this linear sugar molecule. So the synthesis of this sugar molecule is done by this enzyme, starch synthase. It turns out that it has two sites where the reaction happens, and the sugar molecule is activated as ADP glucose.

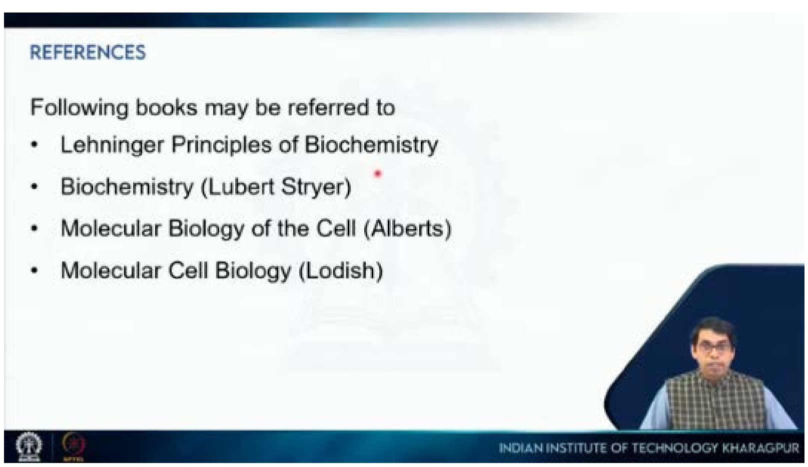
So this is your glucose, a six-carbon sugar, and to it, ADP is added, which is a very good leaving group. So each of the two reactive groups, $X_a$ and $X_b$, at the site of the starch synthase makes a nucleophilic attack on the ADP glucose. So it displaces the ADP, forming a covalent linkage attachment to the C-1 of the glucose subunit. So this attaches the glucose subunit to the C-1. The C-1 and C-4, these are the two sites we are going to look at. So, both are attached to the C-1. Now, the bond holding the glucose residue 1 to $X_a$, this bond, undergoes nucleophilic attack by the OH at C-4 of the glucose residue on 2. So this is the C-4.




So this attacks here and this will attack the next sugar molecule. So what happens then is 2 is attached here after 1, and this 1 shifts here. So $X_b$ 2-1, this is what we get. So 1 is shifted from here to here, and in the position of 1, 3 comes in. Now the reverse attack happens.

So this will attack here. So 2-1 will get shifted here. See 3, 2, 1, and 4 come here. So it will keep on oscillating like this, and the chain will grow on these two sites. So here it is in site B, gets shifted to site A, then again gets shifted to site B. So many repetitions of this sequence extend the oligosaccharide, adding glucose residues at its reducing end.

So this is the end. It will keep on adding more and more sugars at this end and it can go on. So depending on how many reactions happen, you will get a long polysaccharide.



So whatever I have discussed in this lecture, you can find in any biochemistry book, and again, Lehninger Principles of Biochemistry, you can use this as a primary source for studying this. Thank you.