Biochemistry - I

Prof. S. Dasgupta

Department of Chemistry

Indian Institute of Technology, Kharagpur

Lecture # 28
Overview of the Course

In today's lecture we will summarize the course and in doing so we will consider all the functions and the characteristics of the biomolecules that we have studied during the duration of the course. Now initially when we started of I showed you the different topics that we would be covering namely structure and function of biological molecules.

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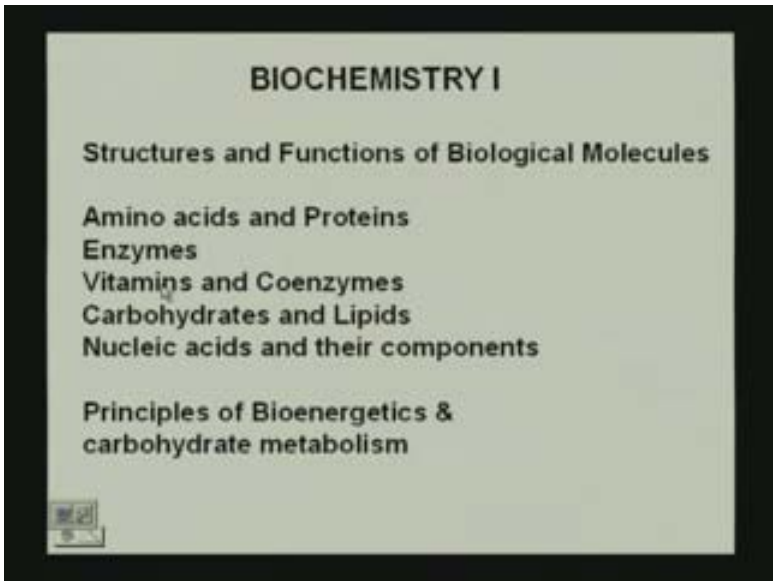

All these molecules biological macro molecules that play an important part in a living system and how they function. Proteins went on to doing enzymes followed by vitamins coenzymes, carbohydrates and lipids and then looking at nucleic acids and their components that is DNA and RNA and what they were comprised of and then specific characteristics which led us on to do the principles of bioenergetics and basically carbohydrate metabolism where we considered the break down of glucose.

In this summary lecture I will be going briefly over all the topics that have been covered here. Now when we consider the biological functions, the basic need is to understand the structure which means that obviously an understanding of a biological macro molecule be it a protein, an enzyme which is a part of a protein which is required for all the specific processes, reactions that go on in the body. You understand that we have to have a

knowledge of its structure because only with the complete understanding of its structure do we know what specific functions it can accomplish.

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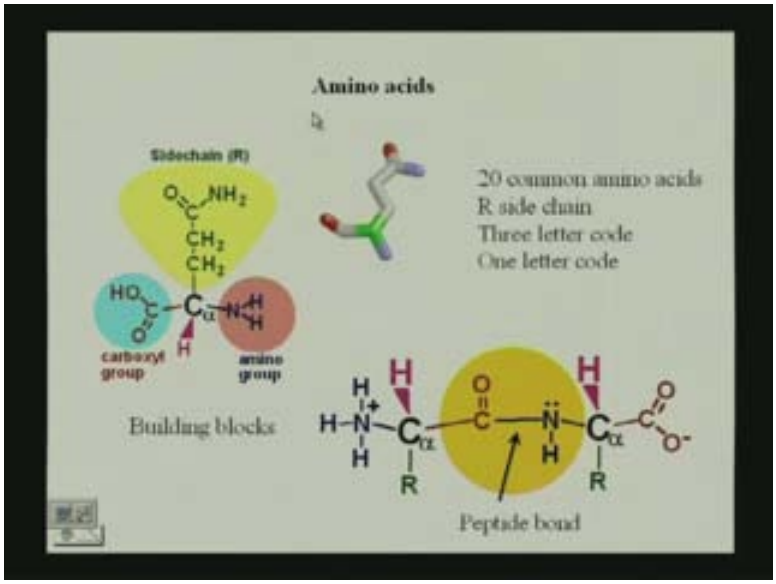

If we look at the basic amino acid structure which is something that you all know by now, we have attached to it an amino group a carboxylic group and a side chain R group which actually is what gives it the specific function, specific characteristics that make it or allow it to perform certain reactions and undergoes certain changes that are going to bring about the reactions that go on in our body, the chemical processes that go on in our body. The linking of the amino acids together gives rise to a peptide bond that is the basics of any protein structure. So here we have a dipeptide that is linked by 2 R groups and we know that these R groups can have a varied number of properties depending on the components present in the R group, they could be negatively charged they could be positively charged.

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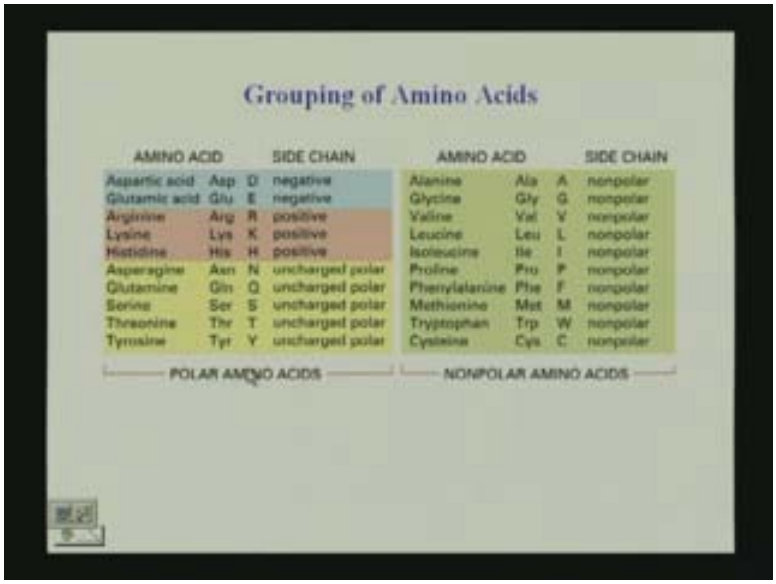

They could be uncharged and yet polar and they also could be in hydrophobic in nature, non polar in nature and we know that on folding of a protein which we will just overview in a minute, we know that these non polar components, the non polar amino acids would likely be at the center of the protein in the core, the hydrophobic core of the protein. In considering the different residues, the amino acid residues we also learnt of their 3 letter codes and 1 letter codes that were used to designate the amino acids linked one after the other, since we know that each of them are going to be linked by a peptide bond.

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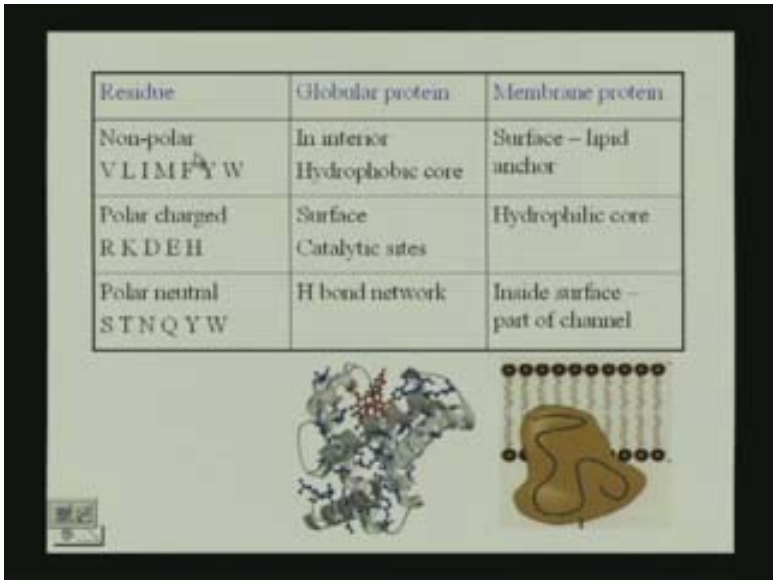

Now in understanding how these residues actually behaved in globular proteins as supposed to membrane proteins, we consider that all these non polar residues the ones that are hydrophobic in nature would in a globular protein rather remain in the hydrophobic core of the structure than on the surface which is the case for the membrane protein. Because when we went on to study the lipid bilayer, we knew that when this integral membrane protein had to traverse the lipid bilayer. It so happened that the core would have to be hydrophilic in nature so that it could transfer the ions but the surface amino acid residues would have to hydrophobic in nature so that they could interact with the lipid fatty acid chains.

However for a globular protein which is interacting with a polar solvent in a polar environment, it is most likely that the polar charged residue would be on the surface of the protein and these would comprise most of the catalytic sites that would bring about the reagents or the substrates to the enzymes for a particular reaction. And the non polar hydrophobic amino acids would prefer to reside in the core of the protein. In looking at the protein architecture we consider the sequence of the protein which is just the linking of the amino acids which is one after the other to form specific secondary structures that are folded amino acid sequences having definite hydrogen bond characteristics, definite areas in what we call the ramachandran plot.

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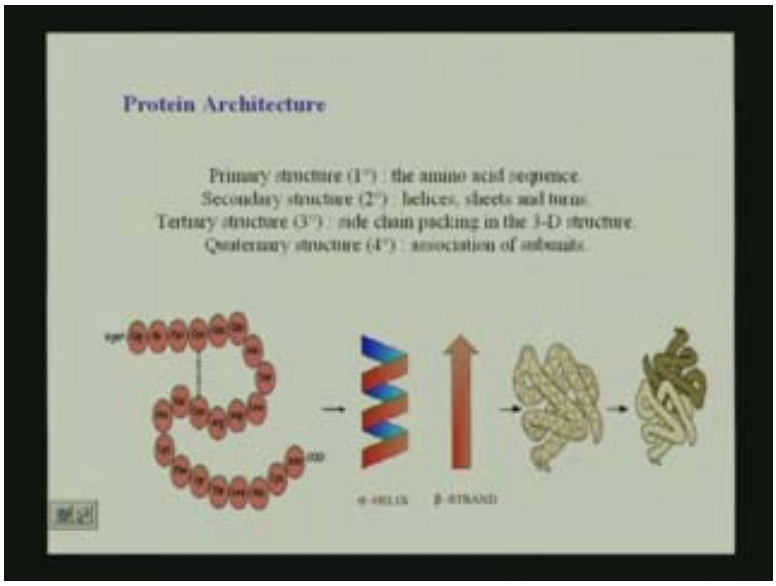

After the formation or the understanding of the secondary structure we consider the tertiary structure of a protein which is the native folded form of the protein and in some cases there may be an association of subunits leading to a quaternary structure of the protein for example like a hemoglobin. In the conformation of flexibility that can occur in proteins due to the rotation about the single bonds the peptide bond we know is partially double bond in character because of the lone pair of electrons on the nitrogen atom.

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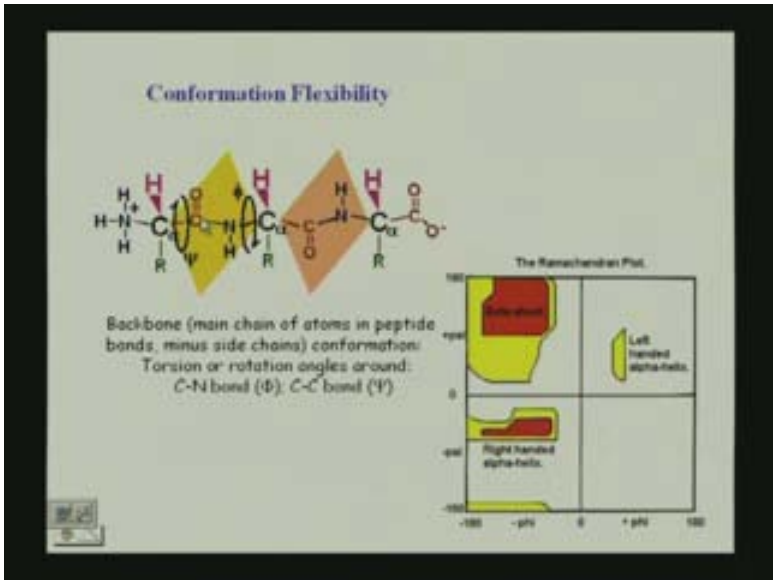

We have a peptide plane that renders this less prone to any flexibility even though we do see sis peptide and mostly we observe trans peptides so that we do not have R groups that may be bulky in nature actually clash with one another. But the rotations are possible about the N C and the C C bonds that give rise to what are called the pi si angles. And we learnt from the ramachandran plot there are definite designated areas for the right handed alpha helix, the left handed alpha helix and beta sheet of the protein which identifies the specific secondary structural elements of the protein on its folding.

Considering other aspects of protein folding this is one very particular example that we consider that is now known as the famous unfinsense experiment where the protein sequence the importance of the protein sequence in understanding the protein structure was emphasized. In this case the ribonuclease a molecule which happens to have 8 sulphide containing amino acids that is 8 cystines that have the possibility of forming 4 disulphide bonds, was actually reduced in fashion where we see that these linkages bring different parts of the amino acid sequence together by virtue of the formation of a disulphide bond.

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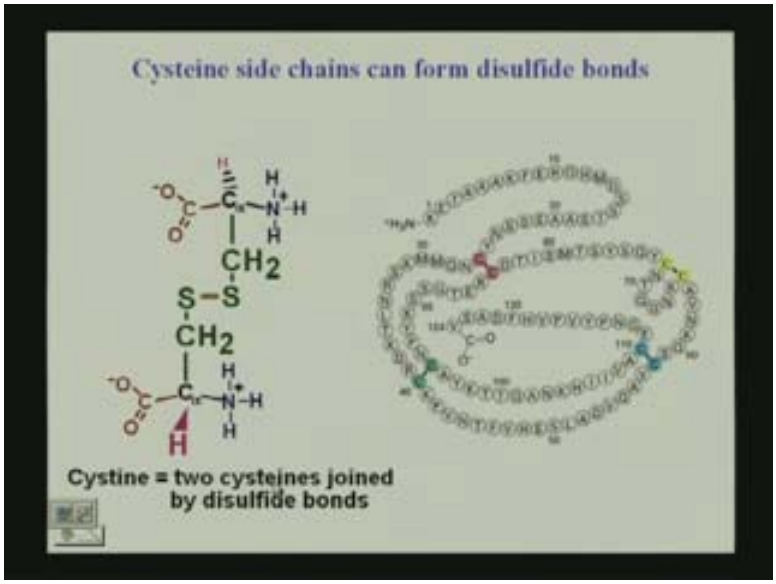

So when we have a cystine formation from 2 cysteines joined by disulphide bonds, we can then break them up with a specific reducing agents. Now if that happens we can actually, in Anfinsen's experiment what was done? Open up this chain to result in it such SH, 8 such cysteines and then allow the protein to fold by removing the reducing agent as well as the denaturing agent and on removal it was found that the exact same bonds or the same disulphide bonds were formed which was actually considered or determined where when the same activity was retained for the protein which meant that it had its final folded native structure.

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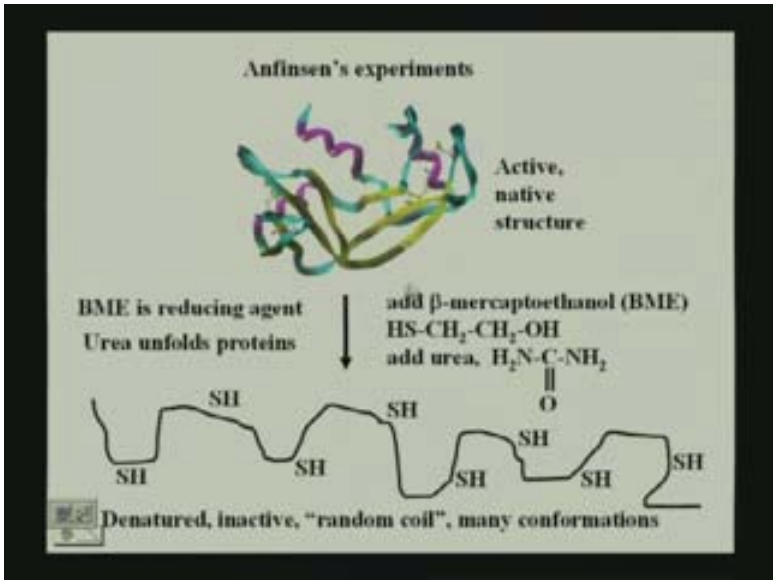

So in this experiment after we have the native structure correctly formed from the 4 disulphide bonds, we know that all the information that is required for the protein to fold is actually present in its amino acid sequence. But this is the protein folding problem where we still cannot predict how a specific sequence goes to this single conformation in its native structure. There are certain methodologies that actually go into understanding the folded protein structure.

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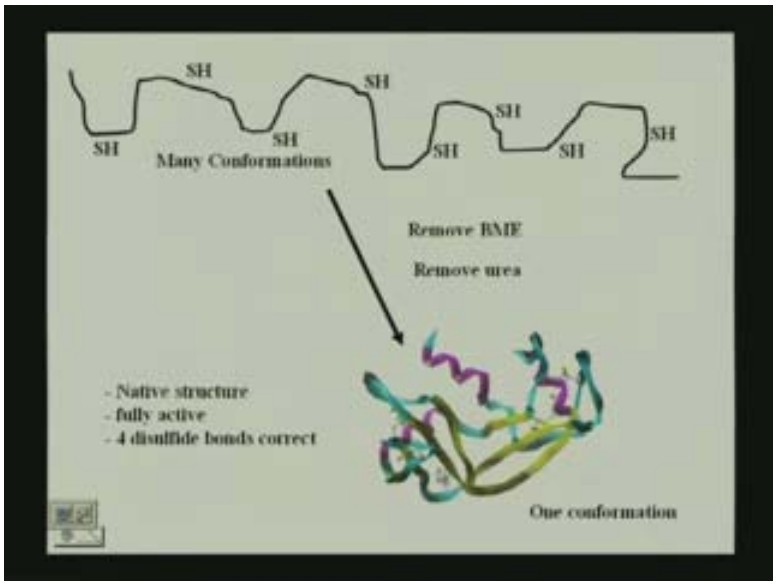

And we know apart from specific guidelines or specific presence of secondary structure where we have hydrogen bonding present there are certain energy terms that can account

or do account for the total energy of a protein and we know that it would rather go to a minimized energy conformation.

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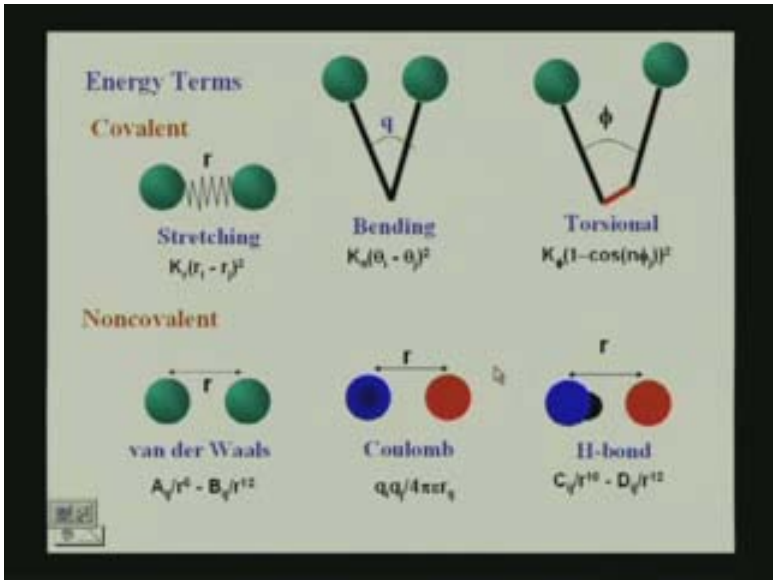

We have covalent interactions and non covalent interactions and the minimization of all these interactions together to give an optimum energy would give us the actual energy or the actual condition that the protein would likely to be in. So we have a stretching energy, a bending energy, a torsional energy that is concerned with linked atoms.

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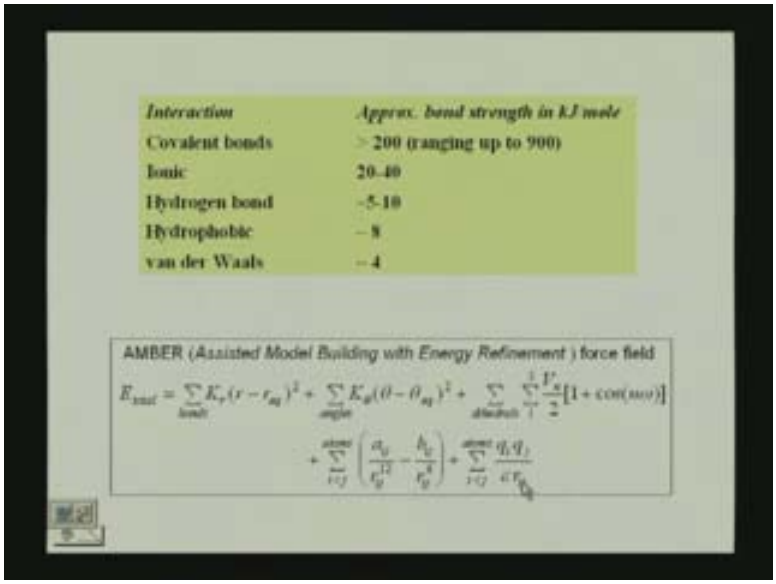

We have non covalent interactions where we have the vanderwaals interaction, coulombs interaction and hydrogen bond interaction together from the different enegertics that are involved in the formation of these specific interactions either covalent or non covalent. We have a specific force field that gives us a total energy of the protein and a minimization of this considering certain variations in the bond length or in the angle or in the dihedrals and certain energetic references with regard to non covalent interactions would give us the total energy of the molecule which actually leads to the energy minimized structure which is what we wanted to predict from the primary sequence of the protein.

So the features of the native state say that there is a well defined 3 D structure and there is a characterized molecular function with this and the idea would be to get from an unfolded structure, a total amino acid sequence to the folded structure. So many proteins actually fold spontaneously to their native structure which results in a very fast process but the predictive methods are still not very accurate into determining what the final structure might be and there also other proteins is called chaperones which actually help certain protein to fold to their native form. They assist the protein in folding in to the native form.

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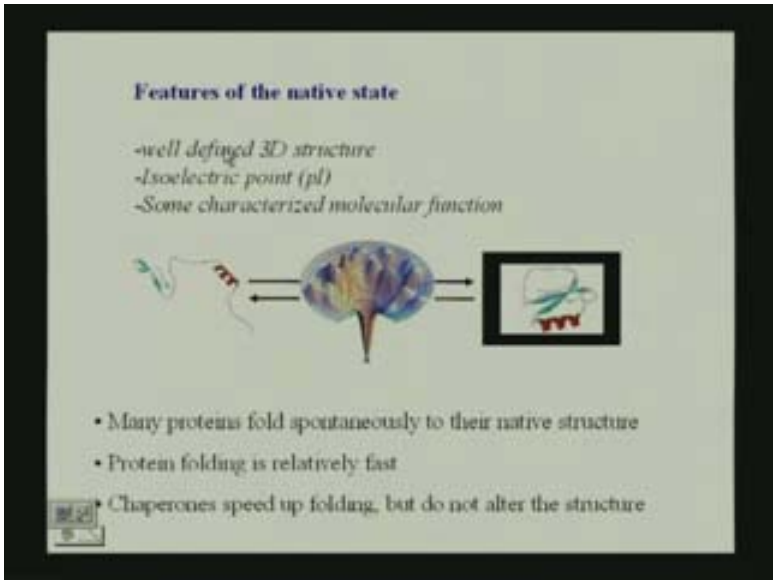

So we consider in the first part of the course where we had our individual amino acid, we considered the various properties of the amino acids and then how they linked up to form the overall protein structure. Now once we have the protein structure or we understand the characteristics of the different side chain residues and what reactions or what types they actually were, then we know what other substrates may interact with them into giving their chemical reactions or their processes or the enzymatic reactions that are extremely important in all biological processes.

So if we consider the thermodynamics of protein folding there are certain aspects of the protein folding that give us what is called a free energy funnel that have a total number of possible conformations leading to a single native structure.

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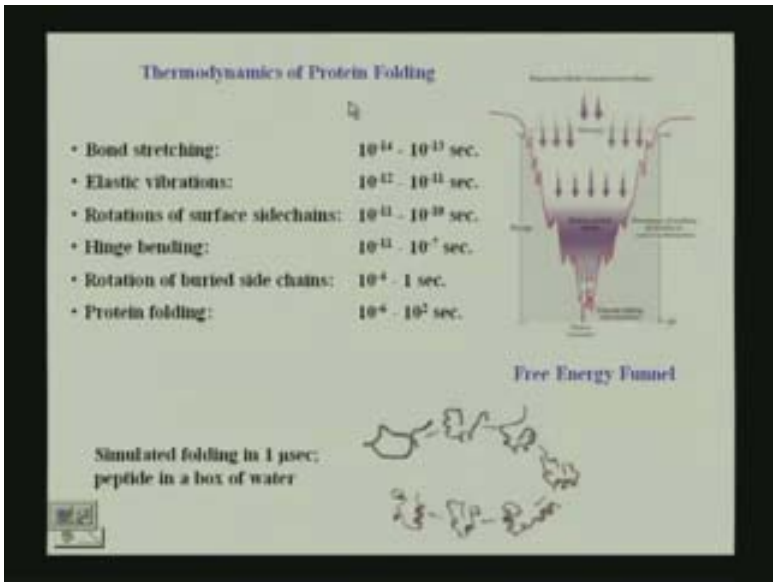

We then went on to understanding enzymes, different types of enzymes we consider that there were 6 classes of these enzymes that actually had specific reactions that they catalyzed because we know that these enzyme catalyst actually reduced the free energy of holding or the free energy of the reaction where we would have had a favoured reaction that would have less activation energy due to the presence of the catalyst which is the enzyme.

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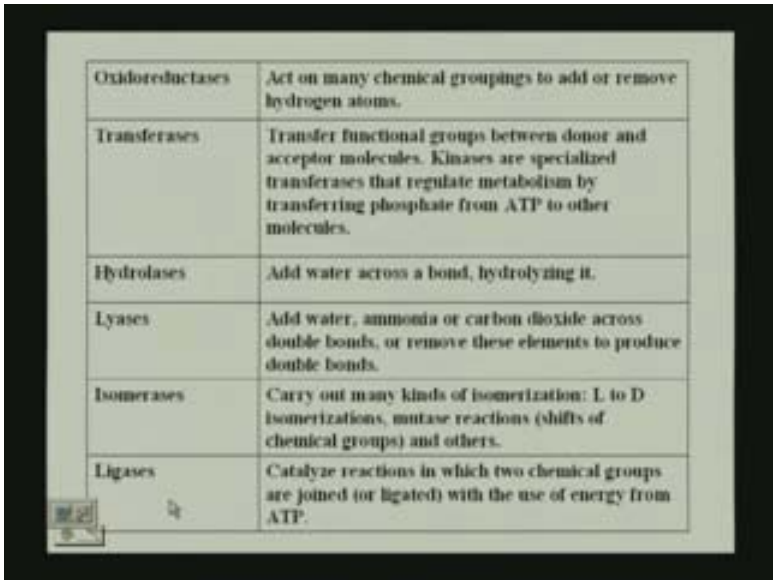

Oxidoreductases	Act on many chemical groupings to add or remove hydrogen atoms.
Transferases	Transfer functional groups between donor and acceptor molecules. Kinases are specialized transferases that regulate metabolism by transferring phosphate from ATP to other molecules.
Hydrolases	Add water across a bond, hydrolyzing it.
Lyases	Add water, ammonia or carbon dioxide across double bonds, or remove these elements to produce double bonds.
Isomerases	Carry out many kinds of isomerization: L to D isomerizations, mutase reactions (shifts of chemical groups) and others.
Ligases	Catalyze reactions in which two chemical groups are joined (or ligated) with the use of energy from ATP.

So the 6 different enzymes possible are oxidoreductase, transferases, hydrolases, lyases, isomerases and ligases. Each of these have their own characteristics there are specific amino acids that are going to be important in bringing about these reactions. For example there are many reactions that we consider called transferases and a specific example of these are kinases that actually transfer phosphate from ATP to other molecules which is an extremely important process in the metabolism or metabolites parts.

Now if we consider the amino acid residues that actually can or do form different forms in the sense that they can act as an acid or as a base and in that case they may be a proton donor or a proton acceptor. Now this becomes extremely important when we consider different enzymatic mechanisms as we had seen when we studied the mechanisms of ribonuclease, lysosime and kimotrpsin which is a protein.

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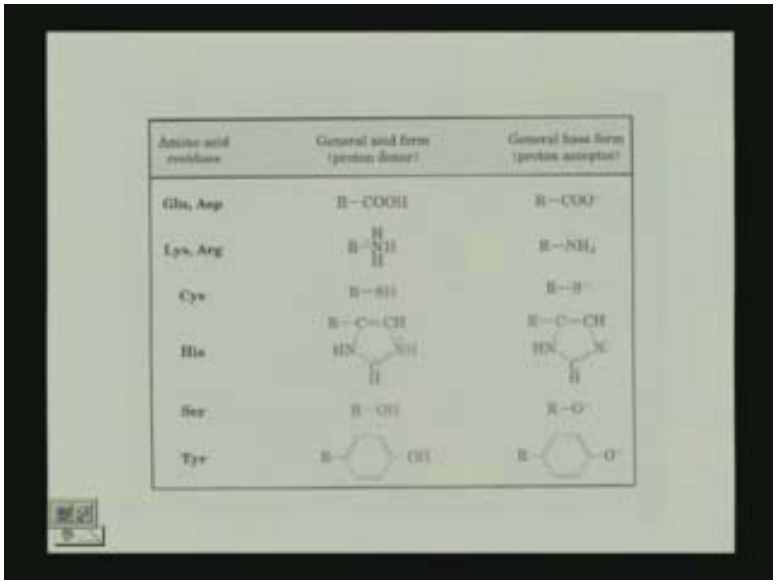

Each of these enzymes have specific amino acids at the catalytic sites and because of their presence and because of their capability of acting as proton donors and proton acceptors, they could bring about certain reactions in the substrate transforming them to the product in the process the enzyme which acts as a catalyst does gets back to its original form. So that it may accept another substrate to form into the product again. In this process we considered michaelis menten kinetics where we considered the velocity of the reaction with substrate concentration and we know that at low substrate concentration we have a first order reaction with respect to the substrate and as the substrate concentration is increased we have the reaction independent of the substrate concentration becoming zero order with respect to s because the enzyme has a limited number of active sites which can accept the substrate molecule. Now from this we studied the maximal velocity, the V_{max} which is the maximum velocity the maximal rate of the reaction which is actually never achieved in reality.

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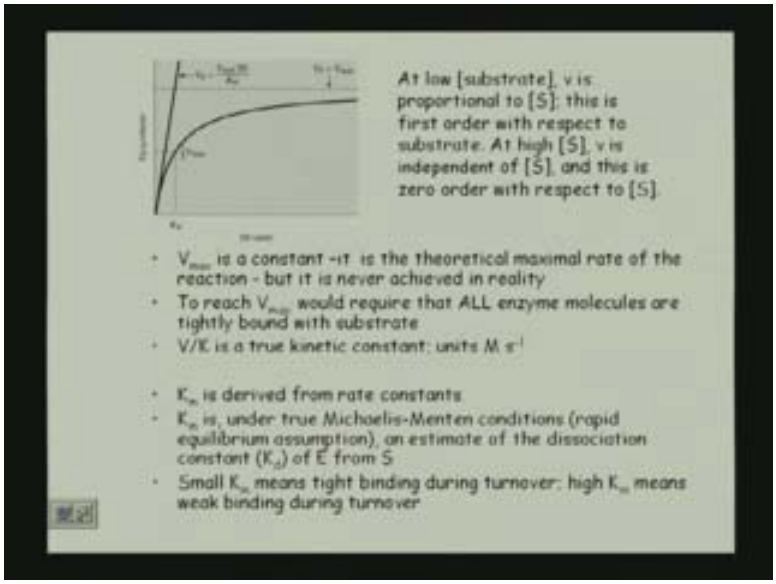

But it is nevertheless useful for specific understanding of the reaction and in this case to reach the V_{max} we understand that all the enzyme molecules have to be involved or tightly bound with the substrate. Then we consider michaelis menten conditions where we derive the michaelis menten constant a k_m value. A k_m value meaning tight binding during the turnover and a high k_m meaning a weak binding during the turnover and we looked at specific expressions which we got from michaelis menten kinetics that actually related the k_m, the substrate concentration and the maximal velocity of the reaction the V_{max}.

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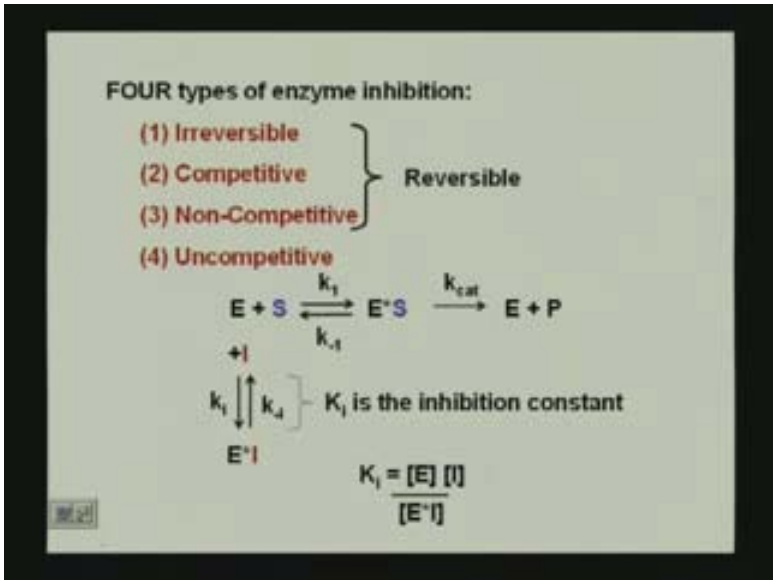

Now considering the V_{max} and the k_m and k_{cat} values, we consider the 4 different types of enzyme inhibition that are possible. We have irreversible inhibition that renders the enzyme inactive for any further reaction. We have competitive, non competitive and uncompetitive and in each of these cases the enzyme or the enzyme substrate complex has some interaction with the inhibitor and we can look at a specific inhibition constant considering the enzyme and inhibition concentrations.

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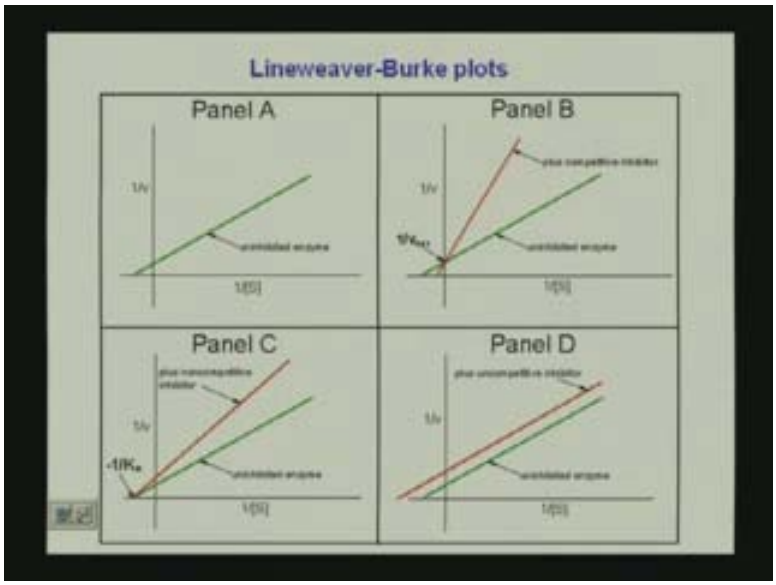

We looked at then line we were work plots that plotted 1 by V versus 1 by S giving us straight lines for the uninhibited enzyme and for specific types of inhibitors, we observe specific types of graphs. The y intercepted this case gave us 1 by Vmax and on the x axis we got the value of minus 1 by K_m for each of the cases which we could consider based on the line we were worked plots for different types of inhibitors, we got a case where for the competitive inhibitor the same V_{max} was achieved. So we knew that this characteristic where we have the same y intercept would mean that this was a competitive inhibitor.

We had a non competitive inhibitor the V_{max} changed but with a sink here and we had uncompetitive inhibitor when we had parallel lines. So this actually summarizes the enzyme kinetic portion from which we consider specific active sites of enzymes, specificities of the enzymes which we studied with relation to ribonuclease, lyzoymes and kimotrpsin.

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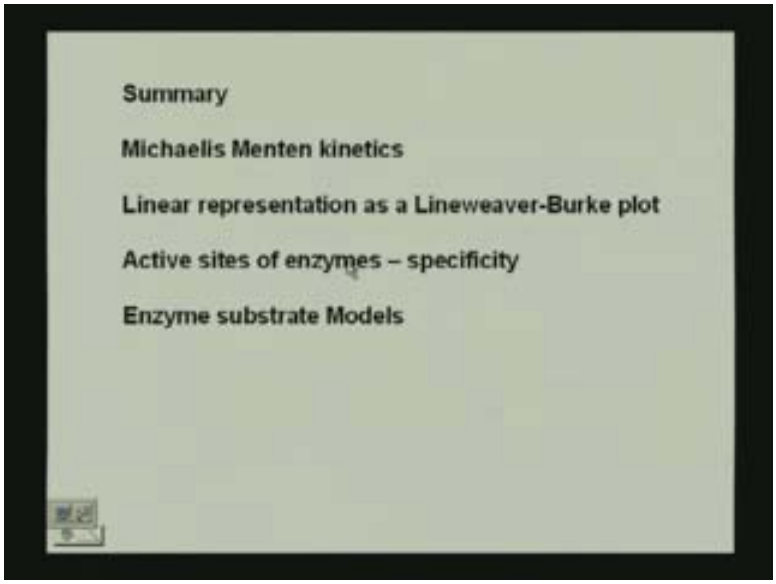

We then went on to do phospholipids considering glycerol. Glycerol when we linked with fatty acids forms the triacylglycerols which were the storage lipids. The membrane lipids that are polar in nature have a part from 2 fatty acids linked to them or phosphate and alcohol linked to it which would be the glycerophospholipids, we have the sphingolipids and we had glycolipids where we had a sugar loop attached to the lipid as well.

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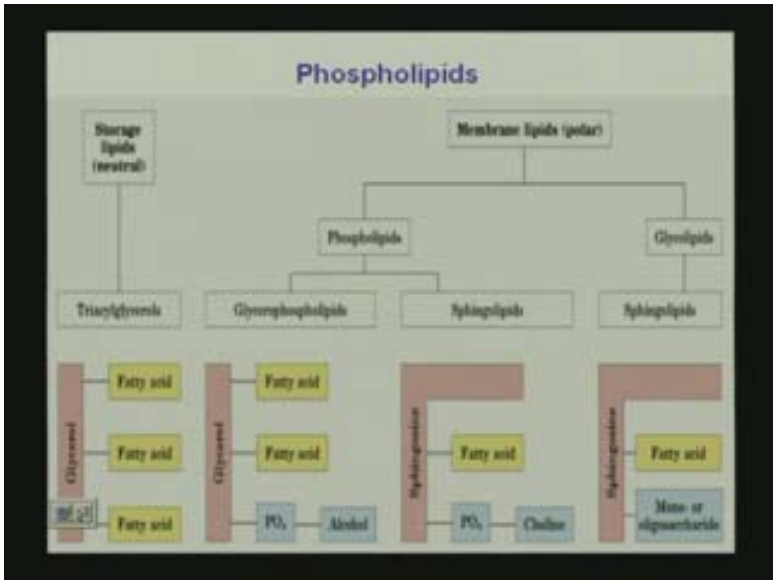

Then we looked at the different molecular arrangements of the lipids that could actually form micelles, liposomes or the bilayer sheet. Now in consideration of the bilayer sheet since this form the cell membrane, we also have transferred across the membrane.

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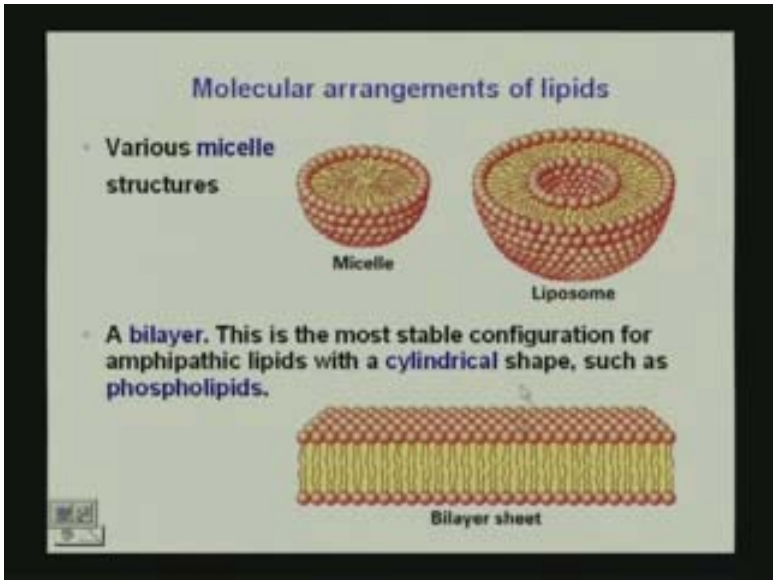

So we studied the specific proteins that were embedded in the membrane where we would have peripheral proteins, integral proteins or once with a lipid anchor. We then went on to consider the fluid mosaic model. The fluid mosaic model actually gave us the idea of how the transportation of material went from the cytosol to the inner part of the say the mitochondria or from the outer part of the cell to the inside of the cell. And we learned there were specific on channel that could actually facilitate the diffusion of ions or other components in and out of a cell and each of the traversing helix we considered were important in their structure in their characteristics.

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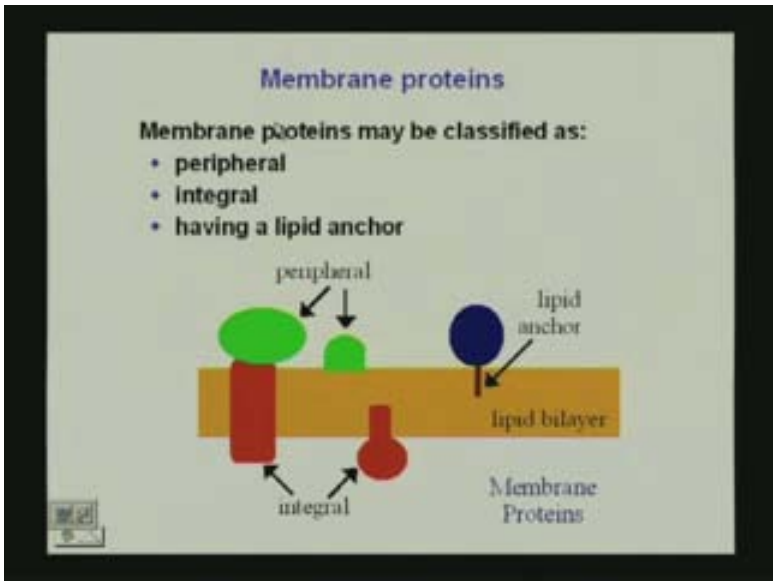

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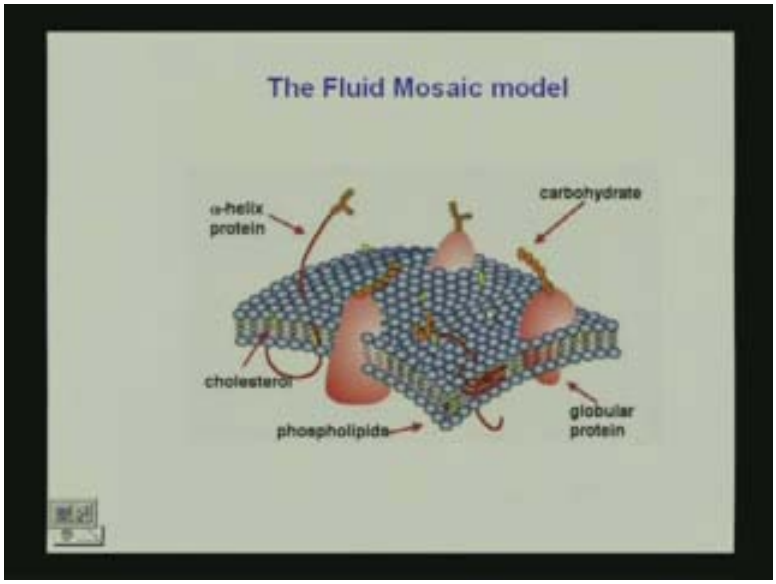

We also found that they were beta porins, porin molecules that actually formed a beta barrel structure that would also account for the transfer of material inside and outside from the extra cellular to the intra cellular or from the intra cellular to the extra cellular and we looked at the specific proteins that were involved in those cases. In considering membrane transport we also looked at the specific electrochemical potential that is involved in the process where we have a concentration gradient along with a membrane that a membrane potential that is associated with the electrochemical potential of transfer.

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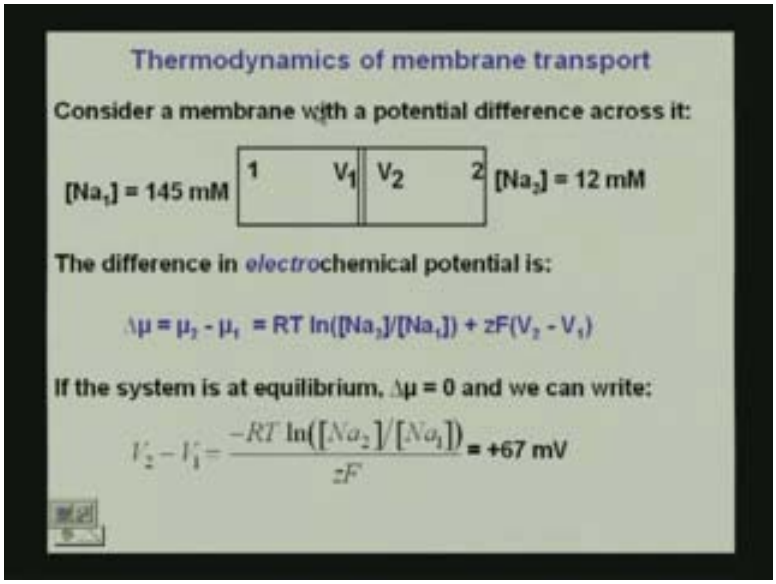

We then went on to study carbohydrates. Now in carbohydrates we consider their importance with respect to them serving as energy stores, fuels and metabolic intermediates that they form as part of the structural framework of RNA and DNA, the sugars and the structural elements in the cell walls of bacteria and plants and in the exoskeleton of arthropods like chitin.

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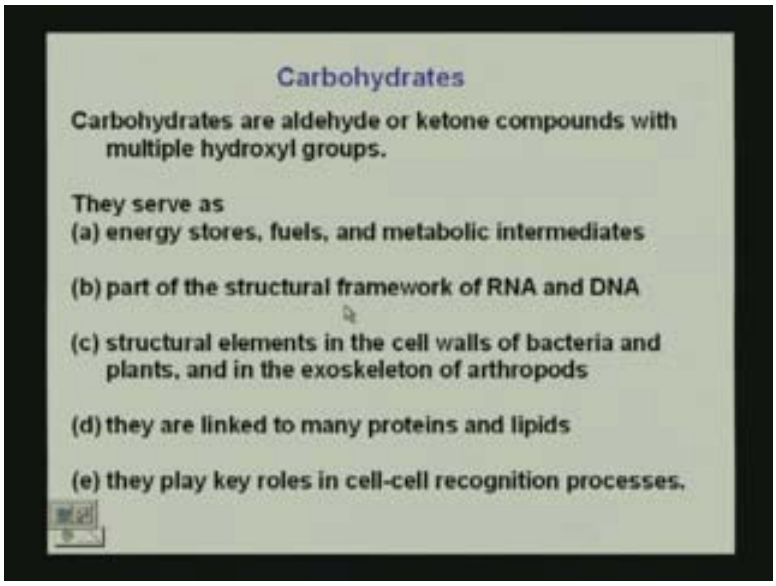

And that they are linked to many proteins and lipids where they form linkages such as these are called glycoproteins and glycolipids and they also play a very key role in cell cell recognition processes. In understanding carbohydrates we consider the monosaccharide that were the simplest carbohydrates and being aldehydes or ketones that have 2 or more hydroxyl groups.

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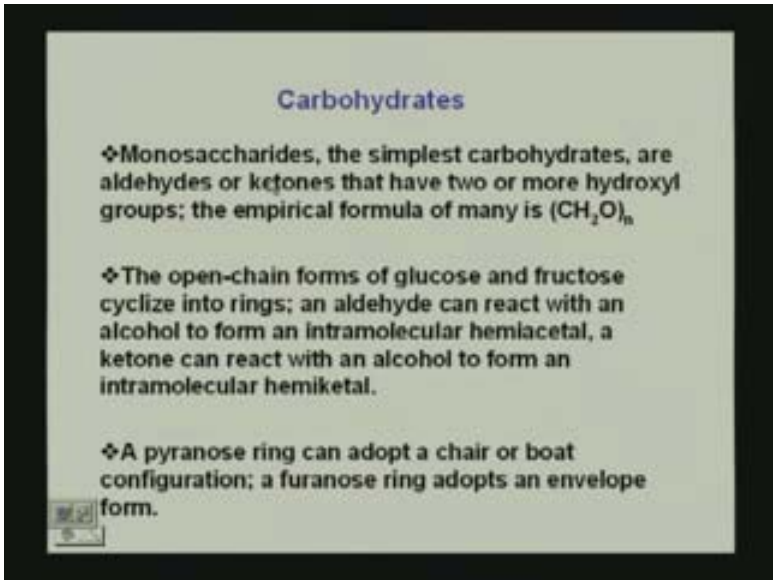

In this case we consider the open chain forms of glucose and fructose that actually cyclize into rings where the aldehyde can react with an alcohol to form an intermolecular hamiacetal and a ketone reacts with an alcohol to form an intermolecular hemiketal. Now in these reactions there are certain other reactions that take place and we also have different conformations possible where we can have a chair or boat configuration and the pyranose ring that adopts and envelope form.

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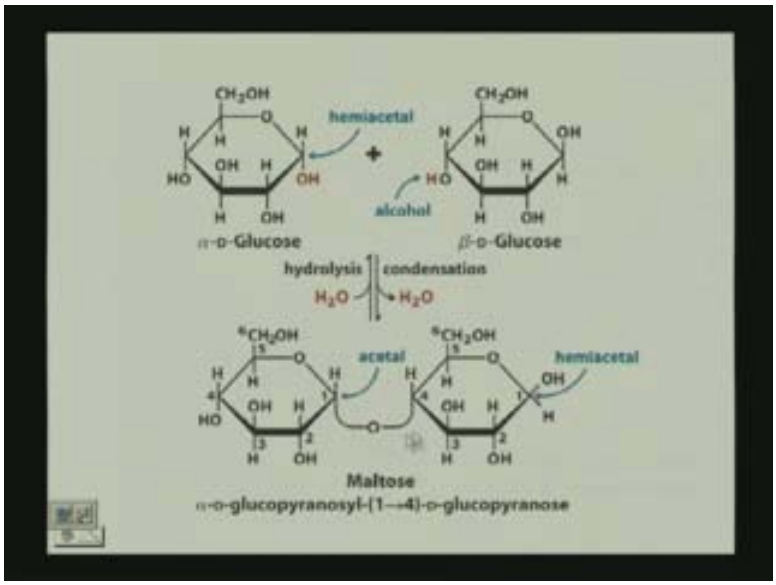

In this case we have the hemiacetal and the alcohol and we have different sort of linkages that link these 2 monosaccharide units to form a specific disaccharide where we have an alpha or beta linkage. We went on to study these alpha glycosidic and the beta glycosidic.

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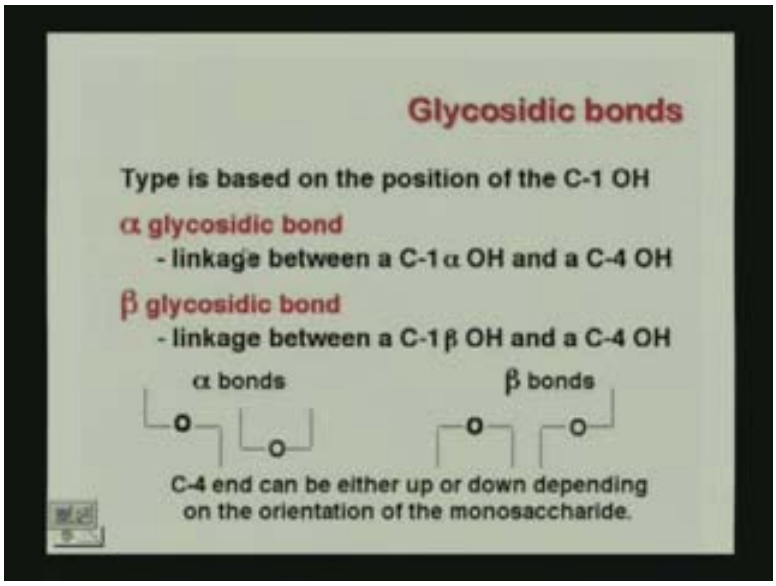

And we learnt from the orientation of the OH, how such a linkage would be possible and based on these linkages how we could have the formation of different disaccharides and which lent to various polysaccharides where we could have straight chain polysaccharides, we could have branched chain polysaccharides and depending on type of polysaccharides that were formed they would have specific functions.

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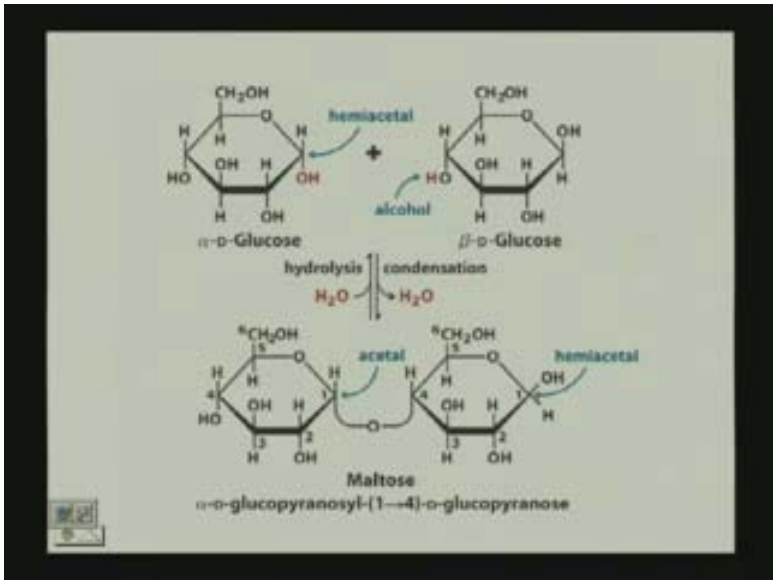

We could have cellulose that would be your fiber type polysaccharide or we could have granular like starch where we would have a granular polysaccharide and each of these were dependant on the type of linkages that linked the monosaccharide units together.

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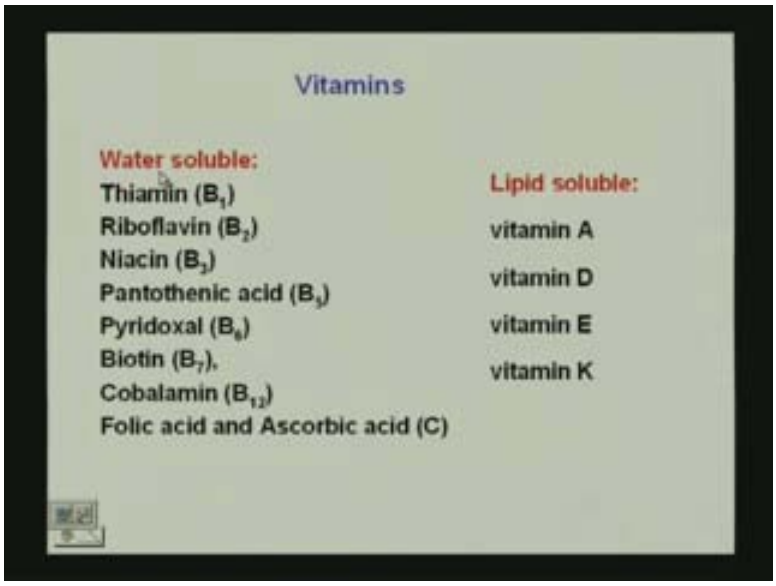

So we then went on to study vitamins. As we went on to vitamins we gradually get an idea of how complicated these molecules can actually get in the biological systems and we looked at a specific series of water soluble vitamins and the lipid soluble vitamins as well. Now each of these vitamins have a specific role to play in the formation of coenzymes. Now in the formation of the specific coenzyme, each of these have a specific role in the final reactions that they would go to.

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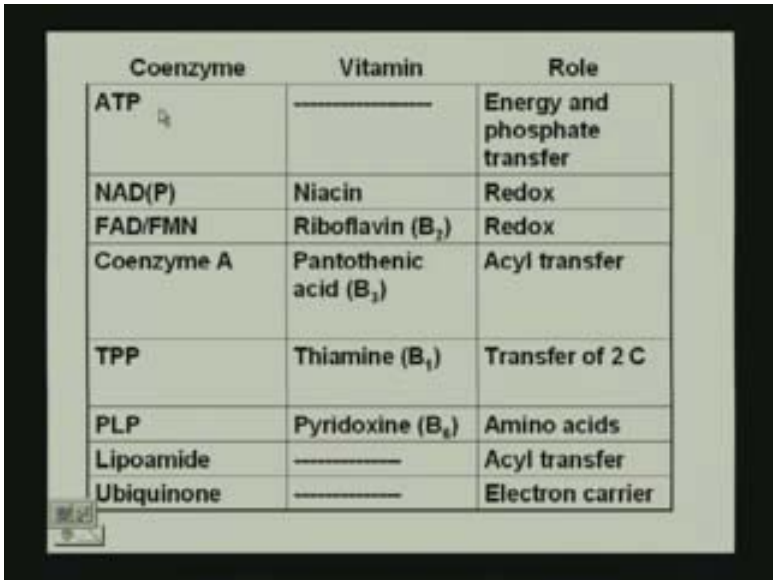

Coenzyme	Vitamin	Role
ATP	----------	Energy and phosphate transfer
NAD(P)	Niacin	Redox
FAD/FMN	Riboflavin (B_2)	Redox
Coenzyme A	Pantothenic acid (B_3)	Acyl transfer
TPP	Thiamine (B_1)	Transfer of 2 C
PLP	Pyridoxine (B_6)	Amino acids
Lipoamide	----------	Acyl transfer
Ubiquinone	----------	Electron carrier

For example if we consider the coenzyme ATP, ATP is extremely important in the specific reactions that it goes by because of the fact that it has a very high energy phosphate bond. In considering this high energy phosphate bond we know that ATP going to ADP plus P_i can give us an energy of minus 30.5 kilograms per mole. Now that is extremely important in the driving of certain other reactions that take place in the metabolic processes as we have studied when we considered the glycolysis which was the break down of glucose.

If we consider some of the other vitamins for example niacin, niacin has a role in the formation of NAD and NADP. Now in this formation we have it play an important role in redox reactions. Now why would niacin or why would NAD or NADP would actually play a role in redox reaction? Or why would FAD and FMN play an important role in a redox reaction? The reason being that these can act as proton and electron acceptors and donors. So we then have an oxidized form and a reduced form. In this reduced form when they accept the hydrogen's. There are certain reactions that require a redox change and in these certain cofactors or these certain coenzymes that are going to be involved in these enzymatic reactions which result in a redox process going on in the body. So we have redox reactions that are usually accomplished by NAD and FAD or FMN.

So considering the type of reaction that is involved each of these vitamins that form the coenzymes have a specific role to play in the biological processes that actually go on. For example when we considered pantothenic acid that is crucial in the formation of coenzyme A which is extremely important in the acetyl coenzyme A component that is required for the glycolytic pathway that gets into the Krebs cycle later on. So all of these cases where we consider each of these vitamins, the roles are extremely important and the roles being possible, the vitamins being transformed into the coenzymes and each of these coenzymes having a specific role to play in a biological process.

For example when we consider thiamine which form thiamine pyrophosphate TPP which results in the transfer of 2 carbon atoms. When we consider pyridoxine, we have PLP we have lipoamide and ubiquinone again another electron carrier lipoamide as acyl transfer. So each of these roles as we consider the specific examples in bioenergetics and in metabolism later on, we understood how each of these were extremely important for the specific function that they were assigned.

In considering nucleic acids and nucleic acid structure then we considered the sugar phosphate backbone. Now this is the primary structure or the primary idea of how the nucleic acids are formed. The backbone is comprised of sugar and phosphate and we have specific bases attached to the sugars and this sugar and phosphate backbone is different in DNA and RNA. The sugars being different and the type of base attached be different.

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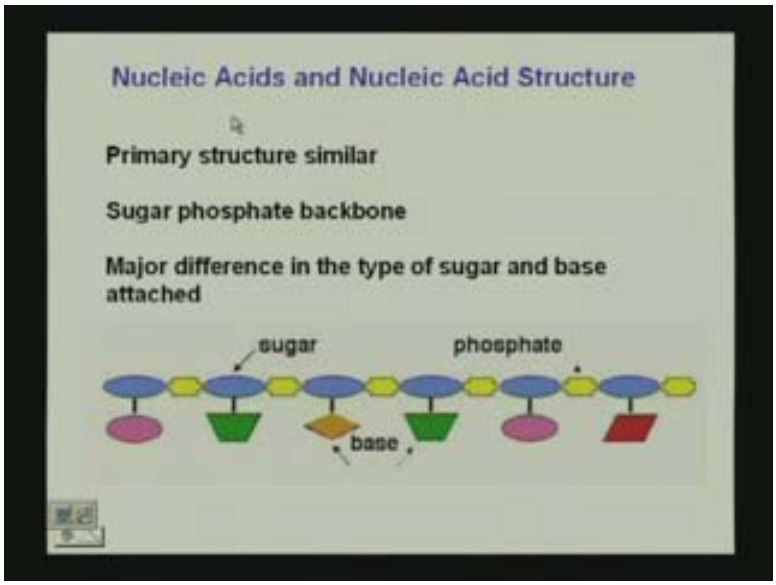

Now the 2 major types of nucleic acids, DNA and RNA, we have specific sugars that were used. Considering the specific sugars that were used we had ribose that were that was used in RNA, deoxyribose used in DNA. Now the difference in this was the specific OH at the 2 prime position that was missing from the DNA sugar molecule.

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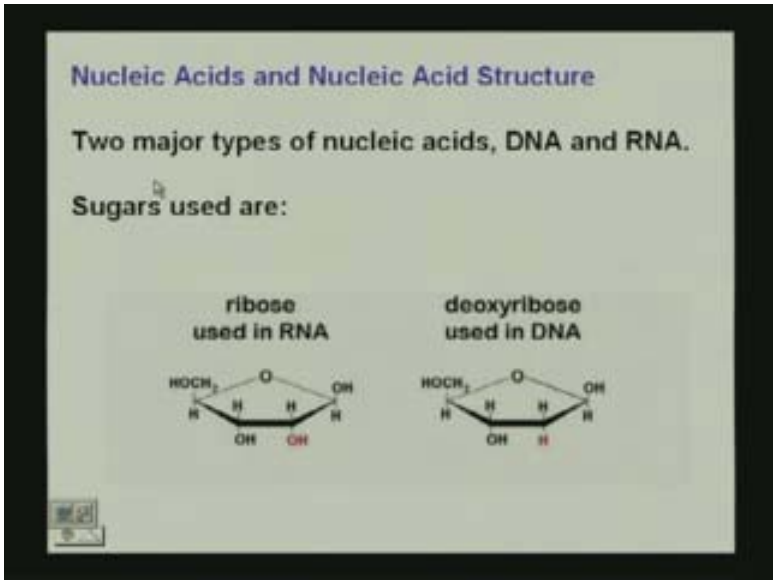

So when we considered these linkages we now know that in case of the absence of the OH group from the 2 prime position, we know that this forms a deoxyribose that is used in DNA and the ribose that is used in iron. The linkages are through the phosphate at this position and we have a base attached to this position. The nucleic acids are made up of polymers, they are extremely important in the specific structure and how actually proteins are formed from nucleic acid structure. We have the central dogma biology which is DNA to RNA to protein.

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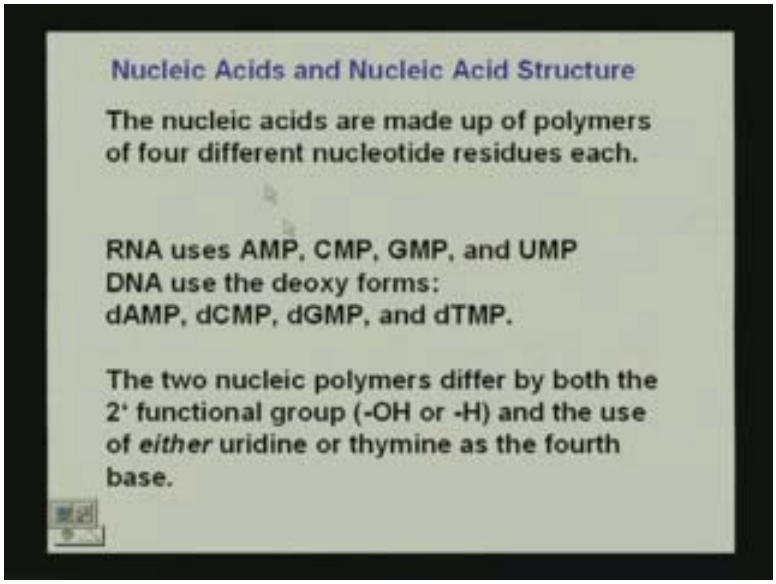

So an understanding of the nucleic acid structure and what comprises their structure is extremely important in understanding how the specific reactions take place or the specific we will look at the basis in a minute right now where the basis actually define which amino acids are going to come one after the other to comprise or to form this primary sequence of the protein of the polypeptide chain and it is this protein that is going to fold into a specific structure, a specific enzymatic structure and bring about a certain reaction. So an understanding of the basic level the base level is important. In RNA we now know that the RNA and the DNA have different sugars that are formed. We have the use of AMP, CMP, GMP and UMP. In DNA we use the deoxy forms of AMP, CMP, GMP and TMP where the small d refers to the fact that this is a deoxy form. And the 2 prime functional group is what is different as well as the uridine or thymine as the fourth base.

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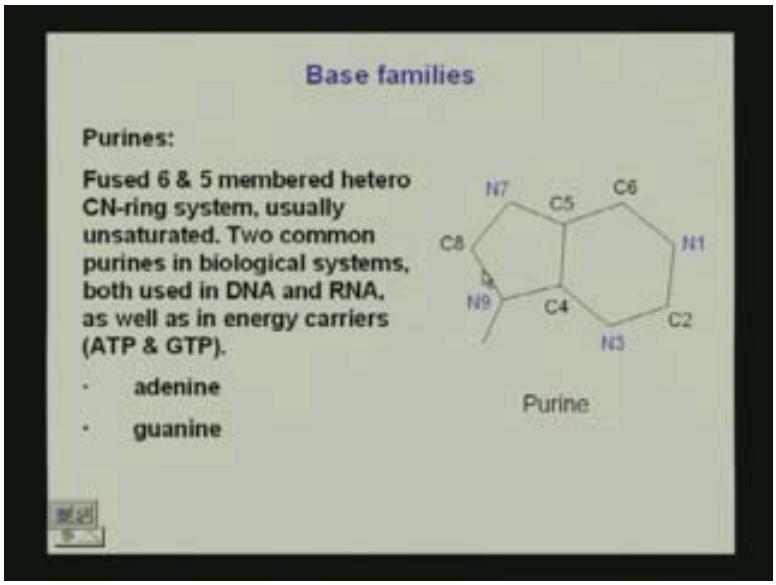

Thymine is what we find in DNA and uridine is what we find in RNA. So what are these guanine, cytosine, uridine and thymine? They are what we call the bases, the nitrogenous bases that comprised of 2 families. The base families that are the purines and the pyrimidines. In considering the purines we have fused 6 and 5 membered hetero CN carbon nitrogen systems that are usually unsaturated and the 2 common purines in biological systems that are used in DNA and RNA are adenine and guanine which are the purine, forming the purine family.

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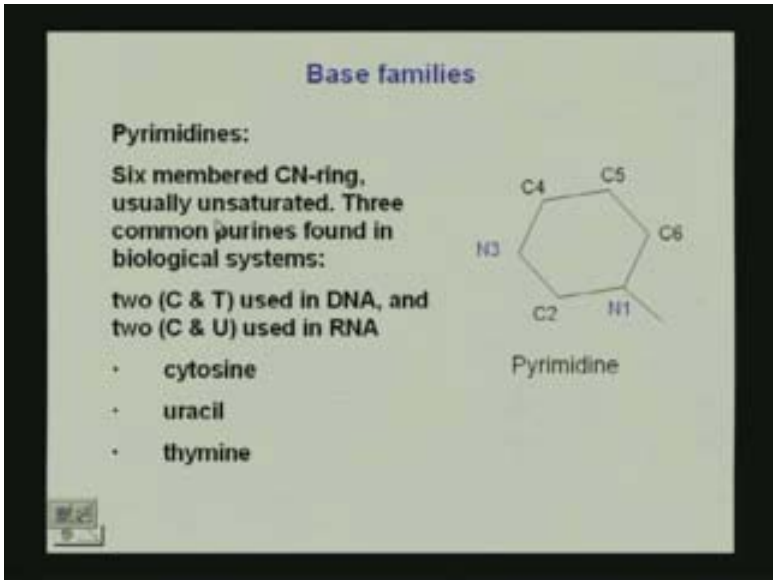

If we consider the pyrimidine family, we have a 6 membered carbon nitrogen ring that is also unsaturated mostly and the three common purines that we see here cytosine and thiamine that is used in DNA and cytosine and uracil that is used in RNA. So if you consider the purines and pyrimidines together, we have adenine, guanine, cytosine, thymine and uracil.

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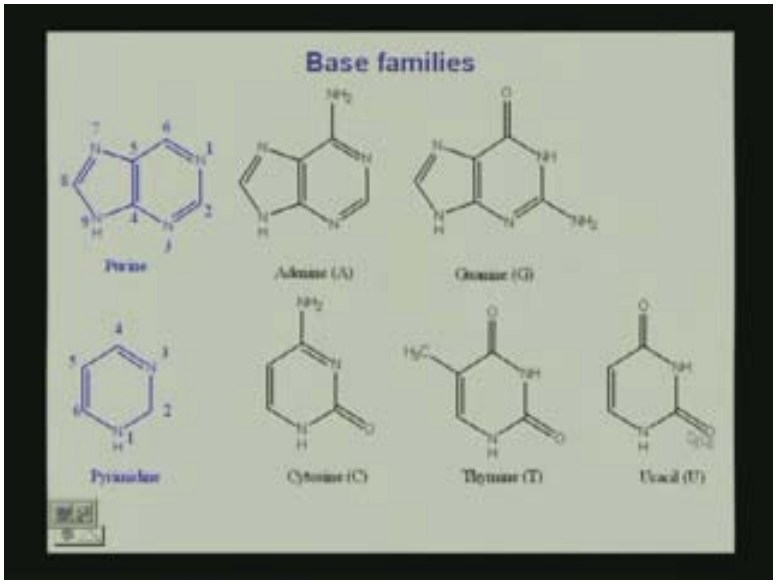

The six membered carbon nitrogen ring being formed from the pyrimidine family and we have the adenine guanine formed from the purine family. And it is the linkage of these to the sugar ring.

The sugar ring at this position that is going to result in different formations and different linkages. These different linkages are different order of bases is going to result in a different form of a protein because a different amino acid codes for every triplet code on as I will just mention again in a moment.

So what we have in DNA is we have the sugar phosphate backbone and all of us remember now that this sugar is lacking the OH group at this position. So we have the sugar phosphate linkage that forms the backbone and we have each of these sugar rings linked to a specific base. The bases belong to either the purine family or the pyrimidine family. The important thing here is the understanding of how the bases linked to one another if we are to understand the double stranded double helical structural DNA. When we have the linkages we have a purine liked with a pyrimidine in each case.

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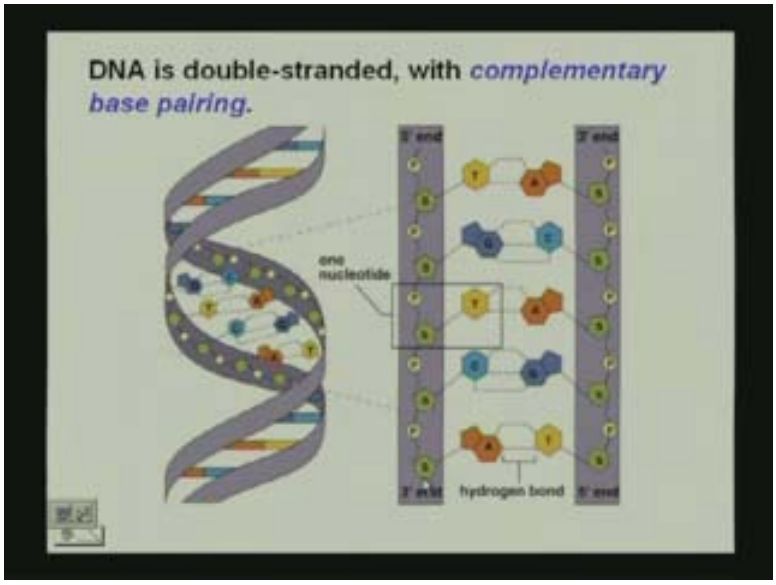

So we can have what is called an AT linkage or we can have a GC linkage. The interesting part of this is that in the formation of a linkage between the pyrimidine and a purine each time, we have a constant distance that we learnt in the classes was around 11 angstroms from one point to the other from the C_1 carbon of say purine in this case to the C_1 of the pyrimidine in this case and that is also 11 angstroms for this particular linkage. So it becomes extremely important in this ladder representation or in the formation of the double helical structure of DNA.

In the formation of the helical structure we have certain interactions that are very important here. So apart from the fact that we have the pyrimidines and the purines linked with one and another where we have one purine linked with a pyrimidine, G linked with C and A linked with T and so on and so forth. We also have specific number of hydrogen bonds in each case, for example when we have an AT linkage we have 2 hydrogen bonds whereas when we have a GC linkage we have 3 hydrogen bonds.

Now what this does is this imparts a certain stability to the DNA depending on the number of hydrogen bonds that we are going to have either 2 or 3 in nature which then leads to specific stability which I will show you in a moment again.

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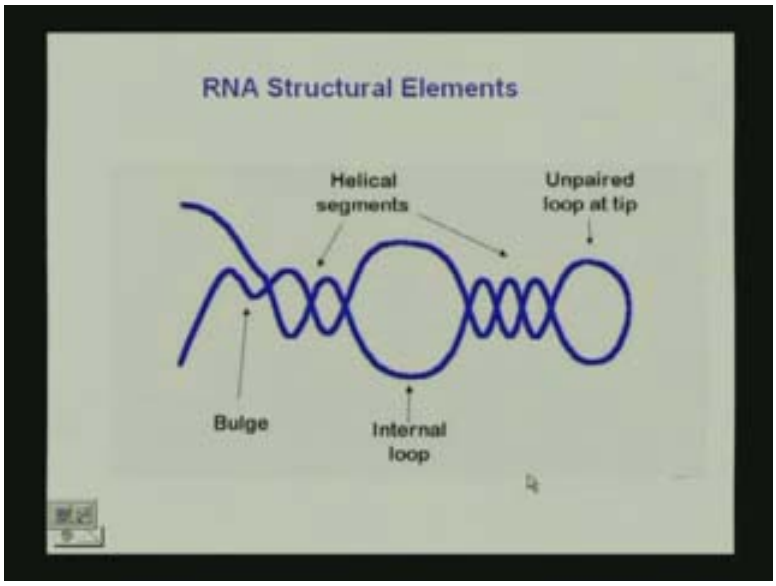

In the RNA structural elements we usually have a single stranded or sometime double helical or an unpaired loop or a bulge because we do not have a specific double helical structure like DNA. But nevertheless there are certain linkages that might also be possible between the purines and the pyrimidines in the formation of hydrogen bonds. So if we compare the structures we have the sugar of the DNA as the deoxyribose, a ribose sugar for RNA.

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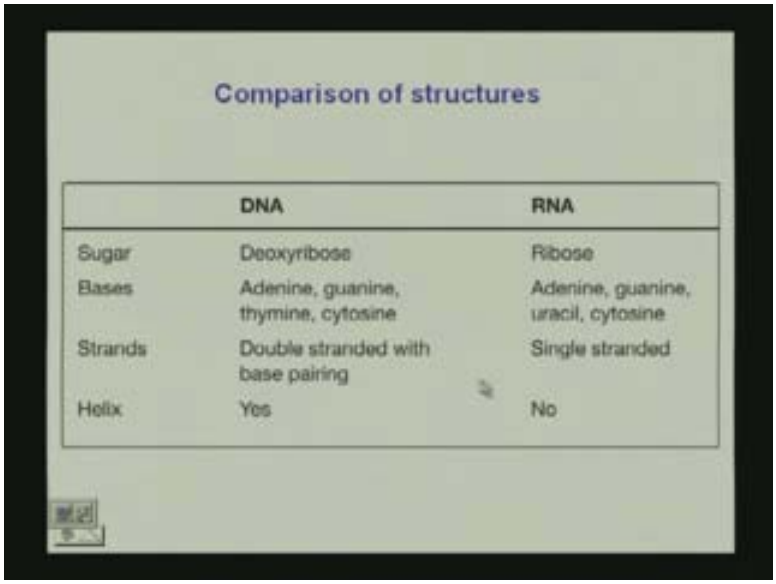

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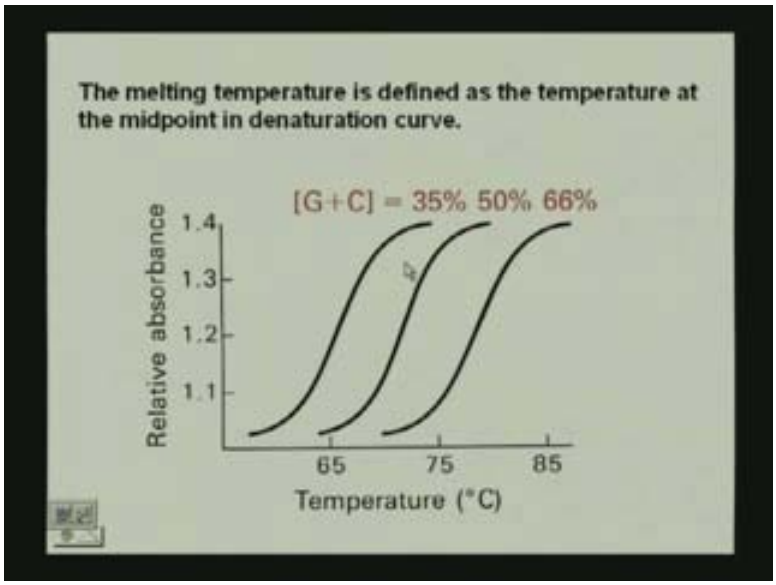

And we looked at specific bases that are possible, the strands and the formation of the helix. If we now look at a specific type, the formation or the presence of a specific number of hydrogen bonds, we can look at what is called the melting temperature of DNA that tells you exactly the percentage of the GC content that you might have which we have certain factors that actually are important in determining what the melting temperature of DNA is. Now in considering these different structures, DNA to RNA to protein there is something else that we also mentioned in class where we considered when we go from DNA to RNA to protein, each of these RNA's they have the messenger

RNA that brings the specific transfer RNA that has the anticodon for a specific amino acid that is going to form a polypeptide chain.

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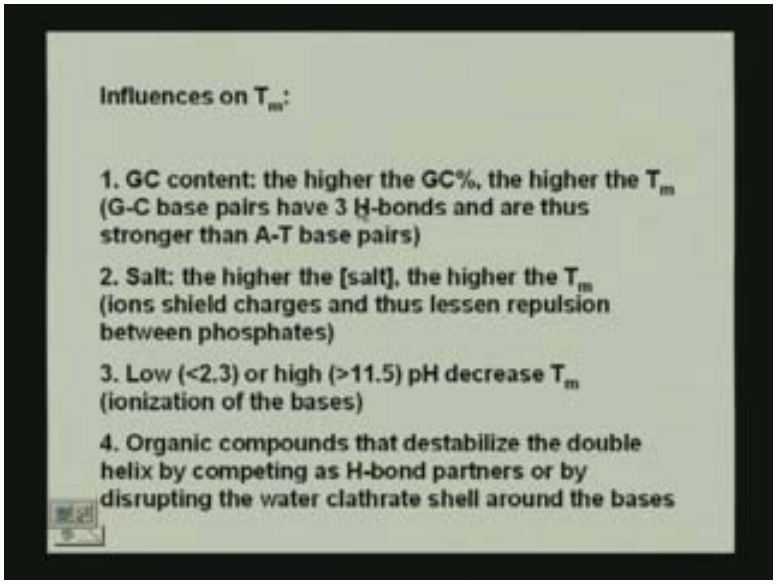

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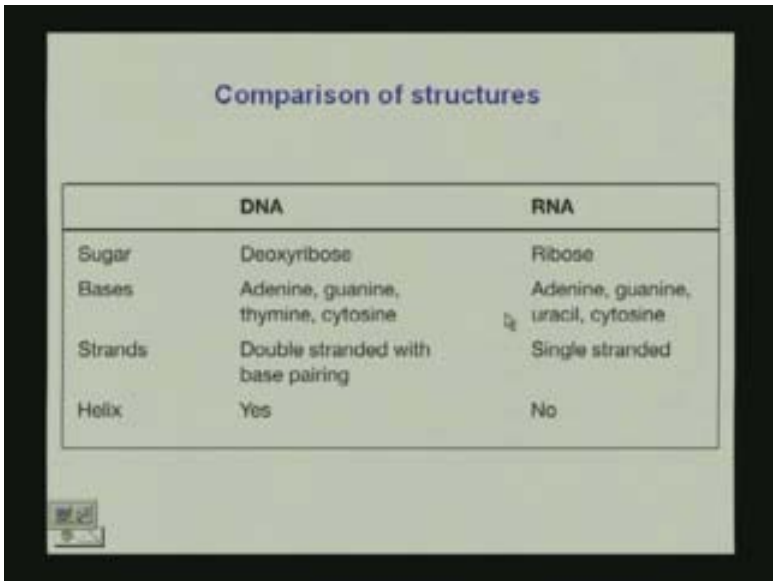

This in the ribosome assembly actually links up one amino acid with another through a peptide bond forming the poly peptide chain. And this folded protein, the polypeptide chain which forms a folded protein is how the proteins are synthesized in the body from the bio synthesis of the protein that is going to eventually give us the enzymes that are responsible for all the reactions. We then went on to the last chapter of the course that involved biochemical and energetics. Now in the process of understanding all the

energetics or all the metabolism of carbohydrates, we consider very important reactions that actually go on in the body.

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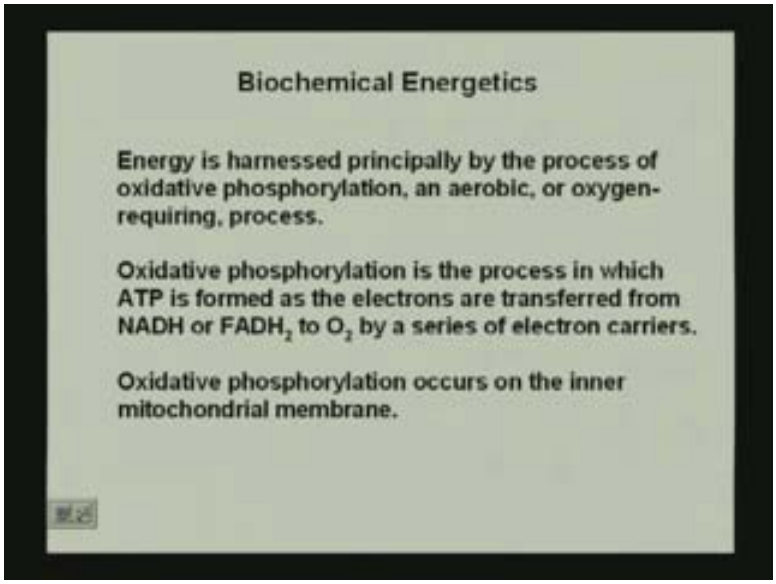

We have the oxidative phosphorylation which is the process in which ATP is formed. Now ATP is the energy currency of the body and it is extremely important that the understanding of this oxidative phosphorylation which is an aerobic oxygen requiring process will actually occur by a series of electron carriers that ultimately transform the electrons to o resulting in the reduction of O_2 to water.

Now in all of the reactions, in energetic reactions that go on in the body, no energy goes to waste. For example if there is a particular reaction and particular reaction that is not required to occur all the time, in that case nature has chosen such a very beautiful way of adjusting this property. When we have the free energy of this particular reaction it is positive making it non spontaneous in nature. But what happens is this reaction is now coupled with another reaction that happens to have a high negative value, a negative delta energy that more than compensates for the positive value required for the specific transformation.

Now as soon as this transformation is required in the body, the ATP or whichever high energy bond is going to come to provide the energy will come into the picture and these two reactions will be coupled together and in the coupling the non spontaneous form will be able to form its product that will then go on to the specific steps of the glycolysis cycle as we will look into the specific example of glucose forming glucose 6 phosphate.

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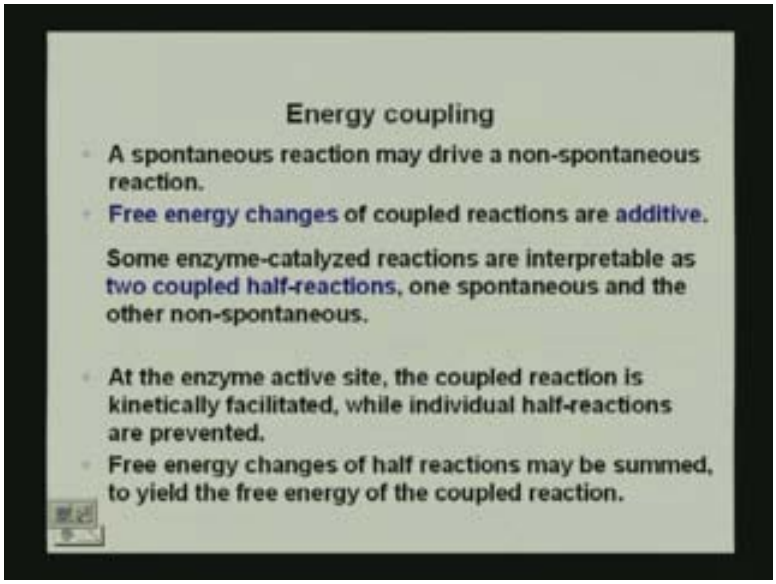

So what happens is since the free energy changes of the coupled reactions are additive, we have two coupled half reactions where one of them is spontaneous and the other one is non spontaneous. Now at the enzyme active site what happens is, the coupled reaction is kinetically facilitated while the individual half reactions are prevented from occurring. For example, if a certain reaction would have a negative free energy then since it is spontaneous it would continually occur but this also does not occur because it is an enzymatic reaction. It has to go to the specific enzymatic site for this release to occur.

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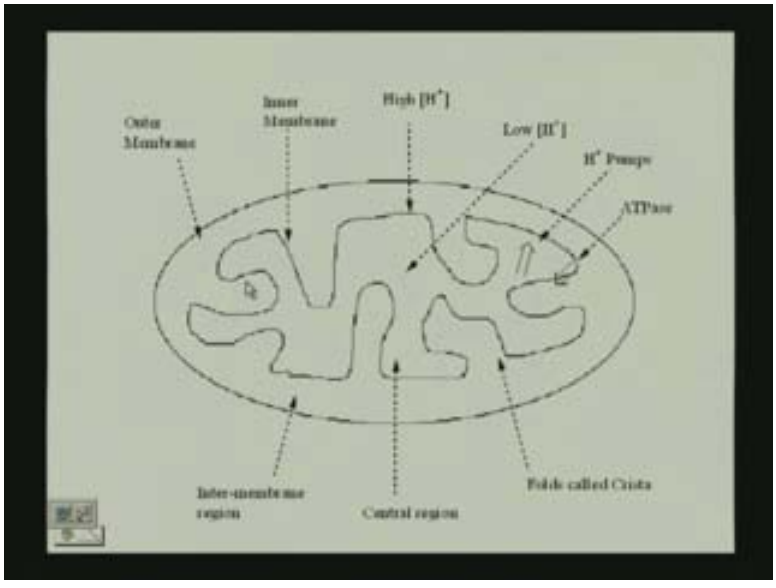

But what happens is the enzymes do not look at these half reactions. They would rather have the couple reaction which is kinetically facilitated but the half reactions which are not. So the free energy changes of the half reactions are summed to yield the free energy of the coupled reaction in various complicated processes that actually go on in the body. We looked at the specific diagram here of the mitochondria where we know that we have proton pumps. We have specific proton concentrations on the inside and the outside, in the inter membrane space and in the inner central region of the protein of the mitochondria rather.

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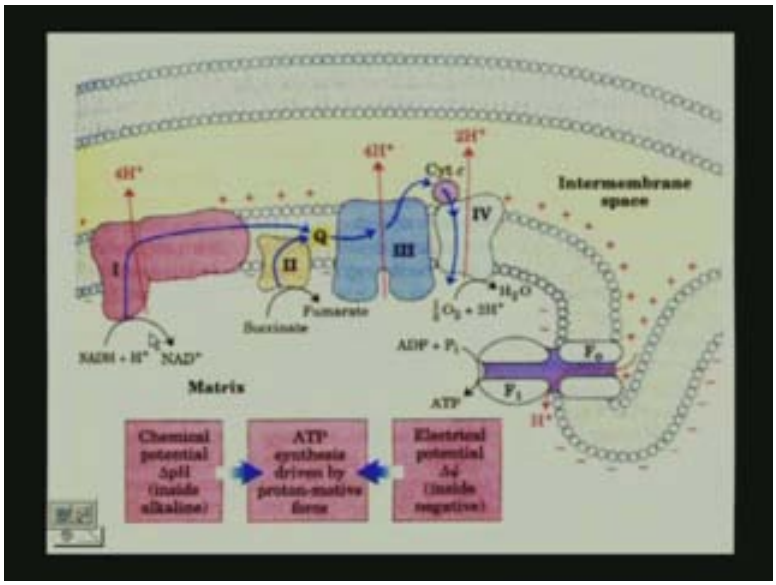

So if we look at the intermembrane region and we look at the outer membrane and the inner membrane, we know that there are specific complexes involved with certain reactions. Each of these complexes actually are comprised of a set of enzymes that bring about certain reactions which ultimately leads to the formation of ATP that is driven by a proton motive force. So what we have is we have a specific chemical potential that arises due to the difference in the hydrogen ions concentrations in these two regions. We also have an electrical potential that is due to the difference in the membrane potential here which results in a proton motive force that ultimately leads to ATP synthesis.

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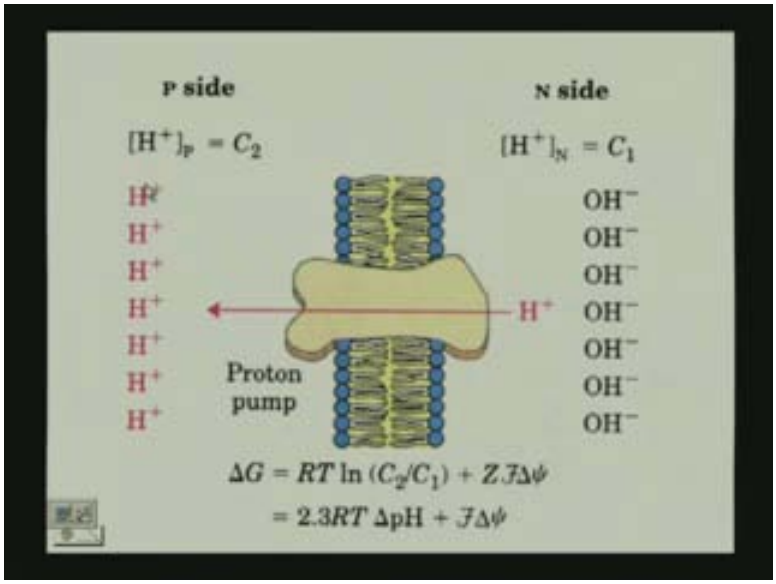

We have therefore a delta G associated with a change in pH because there is a difference in the hydrogen ion concentration. We also have a change associated with a membrane potential, we have the intermembrane space and we have the membrane space and we have a proton pump that actually pumps the protons from this inner space to the intermembrane space which already is high in concentration of protons but nevertheless we need this high concentration of protons to drive the ATPase that is going to result with the protons in the protomotive force to form ATP.

And ATP we know is required in a lot of the reactions that go on in the glycolytic cycle. So what happens here is we have a specific rotation of this top that results in a specific conformational changes of the beta sub units here and the these beta sub units, the catalytic sites will result in the formation of ATP from ADP and P_i.

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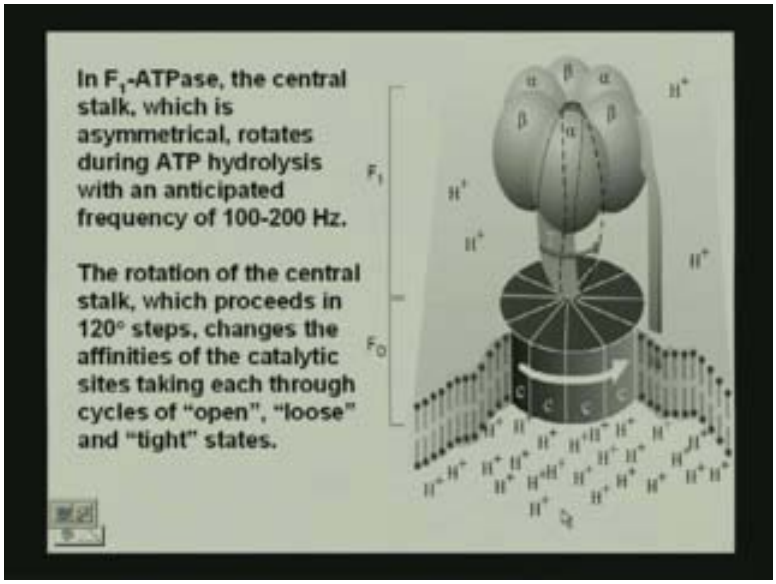

So essentially what happens is we have the formation of ADP plus P_i in a process with electron transport and oxidative phosphorylation to form ATP. In this what happens is we have FAD and NAD plus which actually is formed from the vitamins. We know these are formed from the vitamins, niacin and riboflavin now. And we have protons and electrons come into the picture where we have the reduced form that then with the electron transport forms a oxidized reduced cycle and this is accomplished by a series of enzymes.

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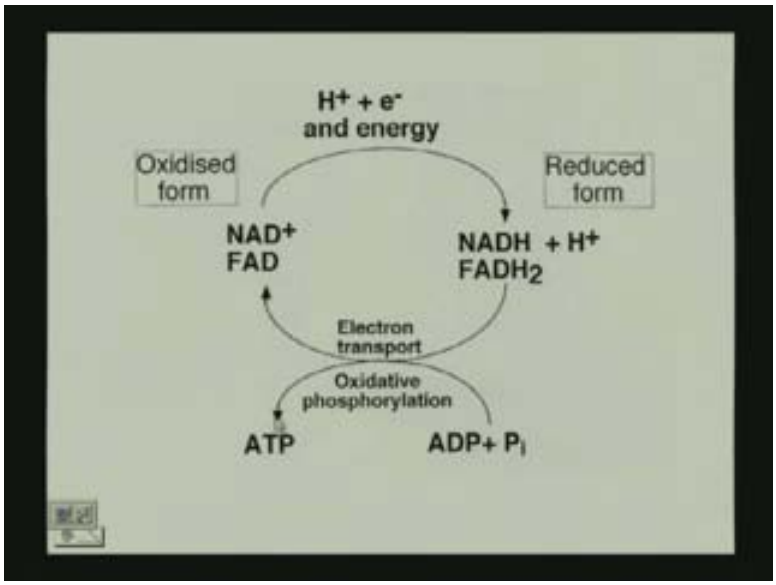

And the overall reactions in oxidative phosphorylation that ultimately lead to the reduction of oxygen to form water in the process it forms ATP. In the formation of ATP

for every NADH that we get, we get approximately 2.5 moles of ATP and from $FADH_2$ we get 1.5 moles.

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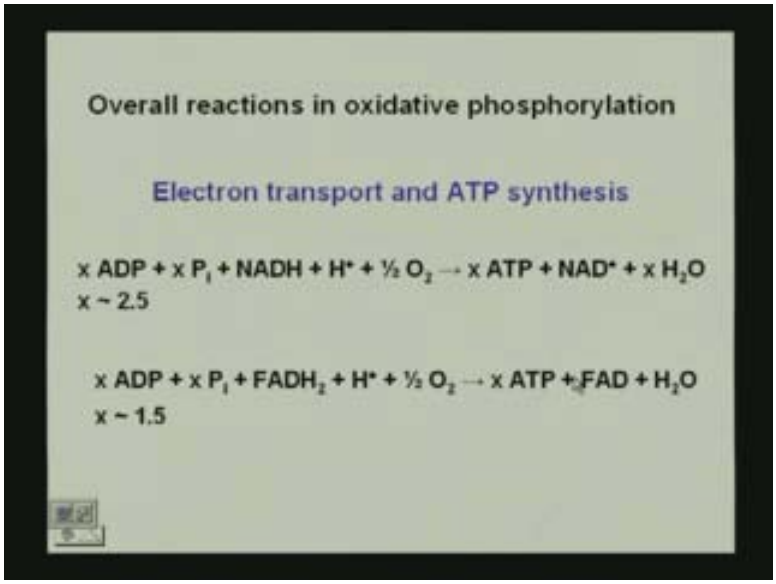

In the catabolic processes that we considered in our metabolism we consider the break down of glucose in this case. But if we look at the overall picture where we our food consumption involves lipids polysaccharides and proteins, each of these break down into smaller units, fatty acids, glycerol. We have fatty acid oxidation glycerol getting into here, then we have polysaccharides that break down into monosaccharides finally get into the glycolysis cycle. We have proteins that break down into amino acids and with further amino acid break down, we finally get into the pyruvate that is the final step of the first cycle, the glycolysis cycle.

The pyruvate then forms acetyl CoA, we learnt coenzyme A formed from phantothenic acid which is a vitamin then from the acetyl CoA it goes into the TCA cycle, offshoot of the TCA cycle being the formation of being oxidative phosphorylation with the utilization of NAD and FAD where we have the formation of ATP and in the glycolysis cycle or the glycolytic cycle also there are steps that involve the production of ATP. The final break down of the glucose is going to lead us into the formation of carbondioxide and water.

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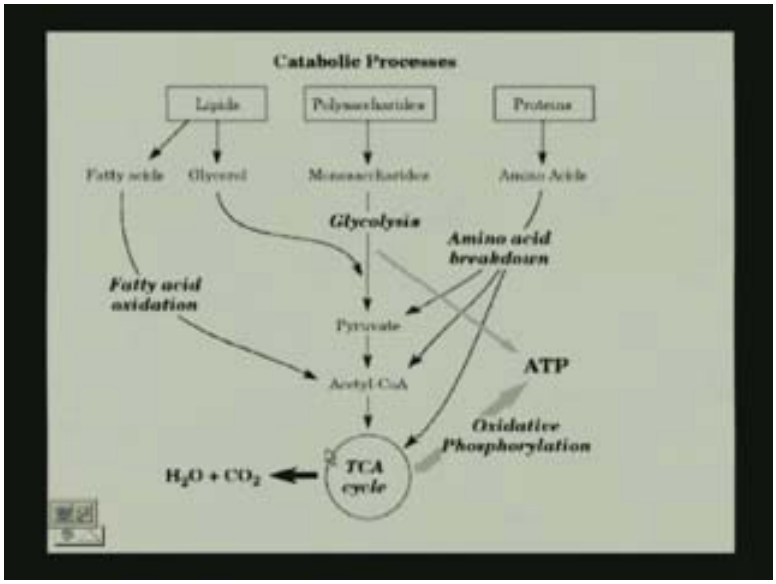

In the glycolysis cycle these are the certain steps that actually go on into this where we are considering the break down of glucose and we learnt in the process of understanding these glycolytic steps. For example in the first step where we have glucose going to glucose 6 phosphate and the delta g associated with this reaction is positive but the break down of ATP to ADP plus P_i results in a large negative delta G. So these reactions are coupled together in the enzymatic site of hexokinase. Hexokinase will then act on these two couple reactions together to give glucose to glucose 6 phosphate and these specific reactions that involve kinase we know, involves a transfer of phosphate myt.

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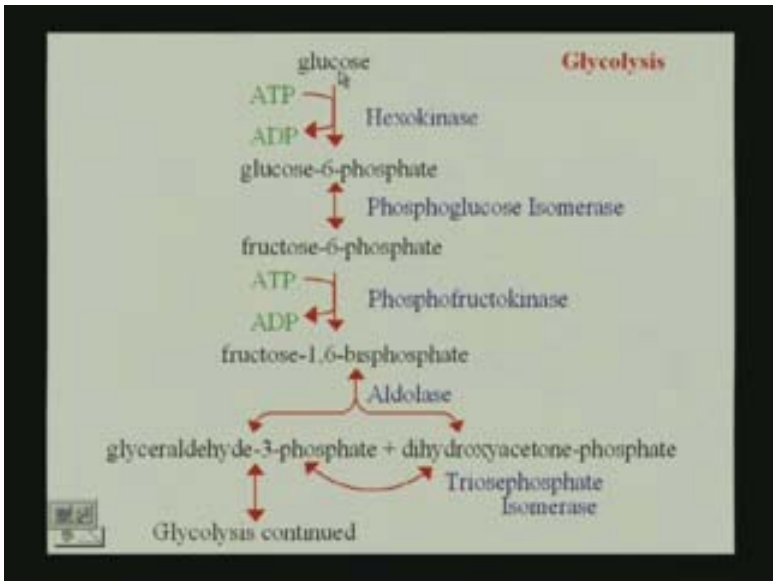

For example the hexokinase or the phosphofructokinase and so on, what happens in these case is these specific steps are irreversible. As a result when glucose is trapped into the cell to forming glucose 6 phosphate it usually does not get back into forming glucose. So once the glucose is trapped in this glycolytic cycle, it will go on into considering the break down of glucose which is finally going to result in our energy formation of carbon dioxide after the TCA cycle. So we have our glucose go to glucose 6 phosphate, this go to fructose 6 phosphate, fructose 1, 6 phosphate where we actually have the breakdown from the 6 carbon unit to 2, 3 carbon units.

Then when we continue this, there are 2 glyceraldehyde 3 phosphate per molecules so this actually occurs twice and this is the step where we have ATP formation and this is another step where we have ATP formation and we have the formation of pyruvate starting from glucose.

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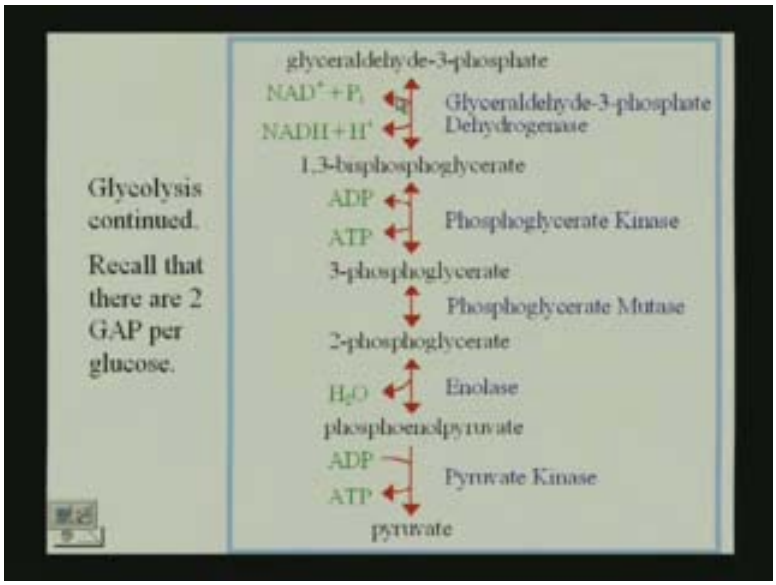

It is this pyruvate that then goes on to the next step so when we have glycolysis, we have this particular step where we have glucose getting into pyruvate. In fermentation we have glucose forming lactate, when the anaerobic catabolism will give us actually 2 high energy bonds of ATP.

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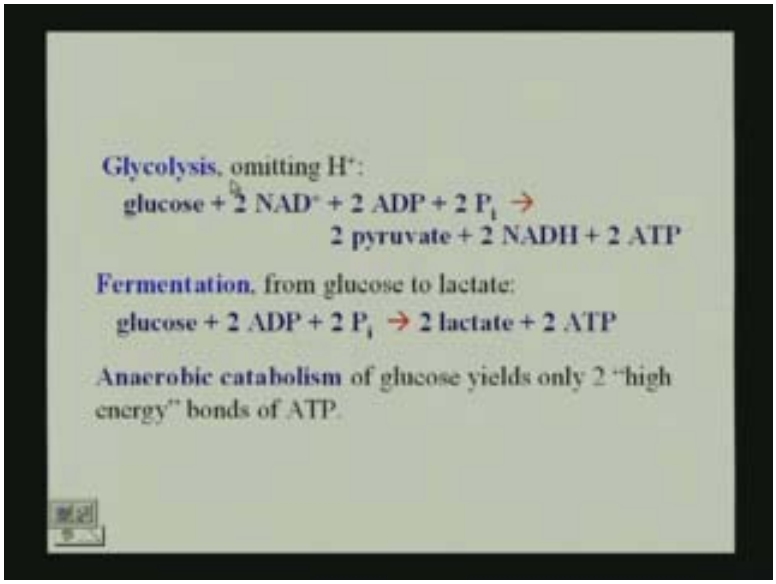

So if we look at the glycolysis enzyme reactions we have hexokinase phosphofructokinase and pyruvate kinase, each of these has a specific delta G_0 prime associated with it, a delta G_0 prime meaning that we are considering the cellular conditions cellular or rather body temperatures into calculating the delta G_0 values where the temperature we consider is not 25 degree centigrade but 37 degree centigrade. So we have our hexokinase, phosphofructokinase and pyruvate kinase that result in negative delta G values and it is these reactions that are actually irreversible in the steps.

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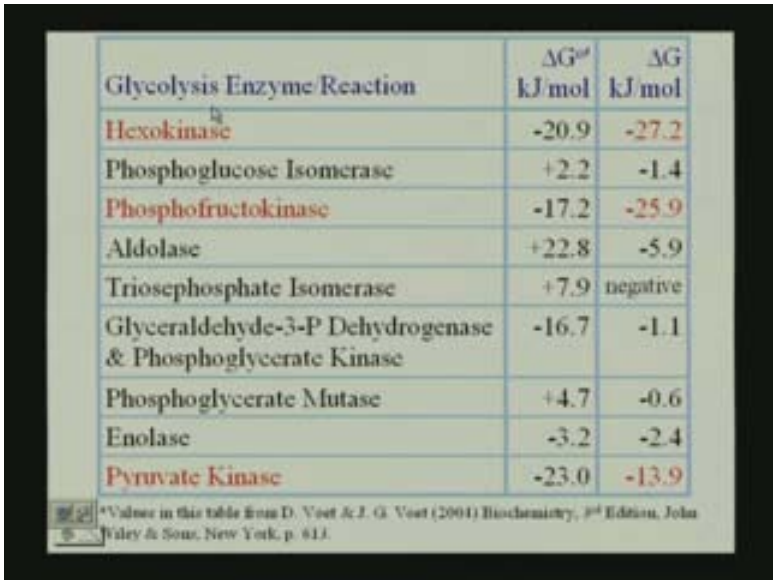

Glycolysis Enzyme/Reaction	$\Delta G^{o\prime}$ kJ/mol	ΔG kJ/mol
Hexokinase	-20.9	-27.2
Phosphoglucose Isomerase	+2.2	-1.4
Phosphofructokinase	-17.2	-25.9
Aldolase	+22.8	-5.9
Triosephosphate Isomerase	+7.9	negative
Glyceraldehyde-3-P Dehydrogenase & Phosphoglycerate Kinase	-16.7	-1.1
Phosphoglycerate Mutase	+4.7	-0.6
Enolase	-3.2	-2.4
Pyruvate Kinase	-23.0	-13.9

*Values in this table from D. Voet & J. G. Voet (2004) Biochemistry, 3rd Edition, John Wiley & Sons, New York, p. 613.

Following this we go for glycolysis, glycolysis is followed by the Krebs cycle. So this is actually what are caused in the cytosol then this is what are caused in the mitochondria.

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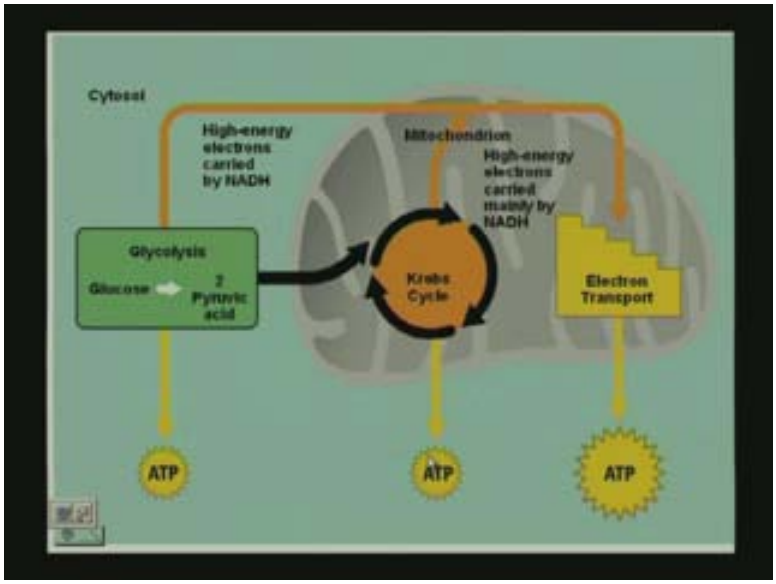

We have glycolysis step, the glucose forming 2 pyruvic acid, we have this pyruvic going into acetyl CoA, then this acetyl CoA leading into the Krebs cycle and after the Krebs cycle the reactions we get from there NAD, NAD plus mainly by NADH the electron transport that finally leads into the formation of ATP. So we are looking at the energy production so the breakdown of glucose is going to give us energy. So we have the Krebs cycle where the acetyl CoA comes in to the picture.

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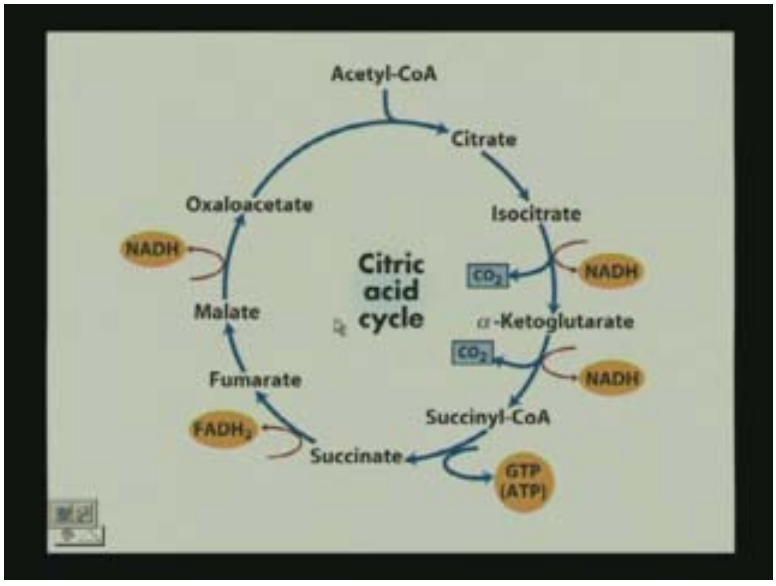

We have the formation of NADH or formation of ATP, formation of $FADH_2$ and NADH which gets on into the electron transfer processes of oxidative phosphorylation. And we have the formation of the carbon dioxide that results from the breakdown of glucose. So if we look at the balance sheet for the phosphate bonds of ATP, we have 2 ATP expended, we have 4 ATP produced so the total net production of 2 phosphate bonds of ATP per glucose and in aerobic mechanisms we have the pyruvate produced in glycolysis that is oxidized to carbondioxide by the Krebs cycle.

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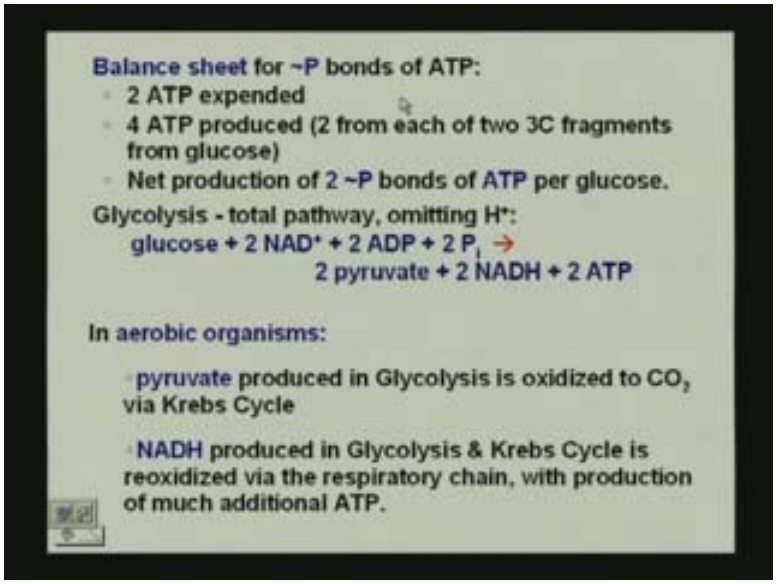

The NADH that is produced in glycolysis and Krebs cycle which we saw in the previous slide where we have the formation of NADH in these steps and the $FADH_2$ that is or rather has to be reoxidized because it is now reduced, it has to be reoxidized. This is reoxidized by the respiratory chain where additional ATP is formed.

So if we look at the total number of ATP that we actually get, ultimately formed from the break down of one glucose, these are the number of ATP formed that is 30 to 32 depending on the specific reactions that we are considering and for each of these 2 cases where we are forming glucose 6 phosphate from glucose or fructose 1, 6 phosphate from fructose 6 phosphate, we require ATP to form the phosphates in a reaction mechanism that involves the enzyme kinase and then we go on to see the number of ATP or reduced coenzymes at a directive form, in each of these steps and we get to a final 30 to 32 ATP ultimately formed from 1 glucose.

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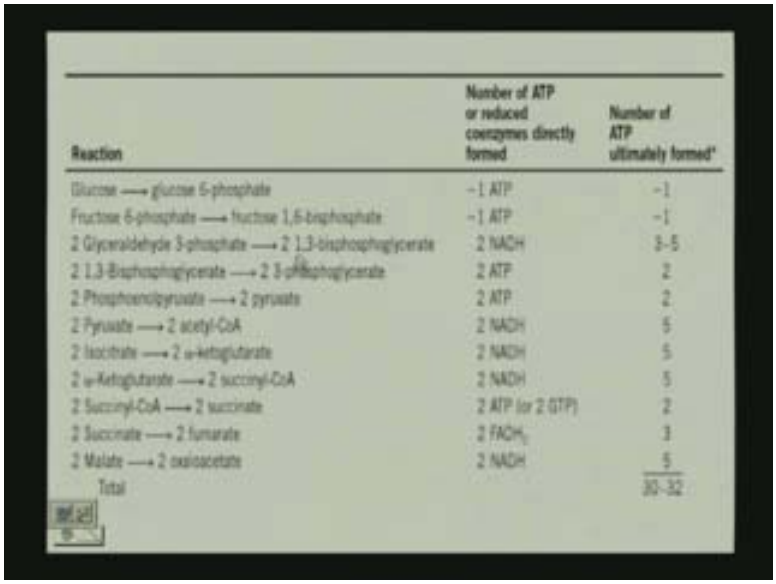

So if we look at the efficiency of biochemical engine in living systems, the oxidation of one glucose actually yields this amount of energy, 2840 kilojoules per mole. So the energy obtained by the biological engine gives us 32 ATP that is 30.5 kilojoules per mole giving us 976 kilojoules per mole and if we consider standard conditions the efficiency that we are going to get is 34% but the cellular conditions are such that it gives us a efficiency close to 65%.

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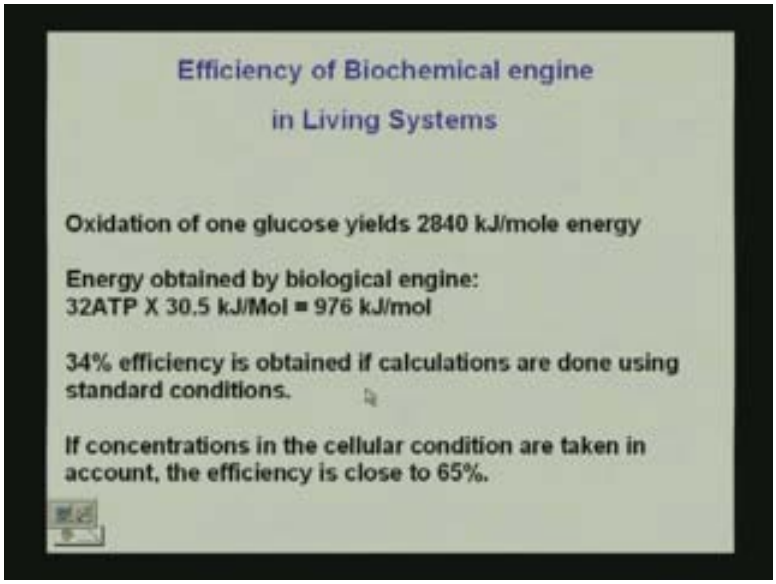

So this is considering the energetics of our system with the breakdown of glucose. So this has been, this course biochemistry one where we have considered starting from amino acids structures of biological macro molecules to enzymes to lipids, carbohydrates, vitamins, coenzymes then nucleic acids components and biochemical energetics this is given you a brief understanding of how biochemical processes or bio biological macro molecules what are present in the body and what their importance is the role that they play in the functions that go on into the body and you have to understand that understanding the structure of each of these components is extremely necessary to get a complete understanding of the function that they can characterize or the functions that they can accomplish or an understanding of the reactions that they can actually accomplish.
Thank you