Bioinorganic Chemistry
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Module - 1
Lecture - 17
Nickel Enzymes

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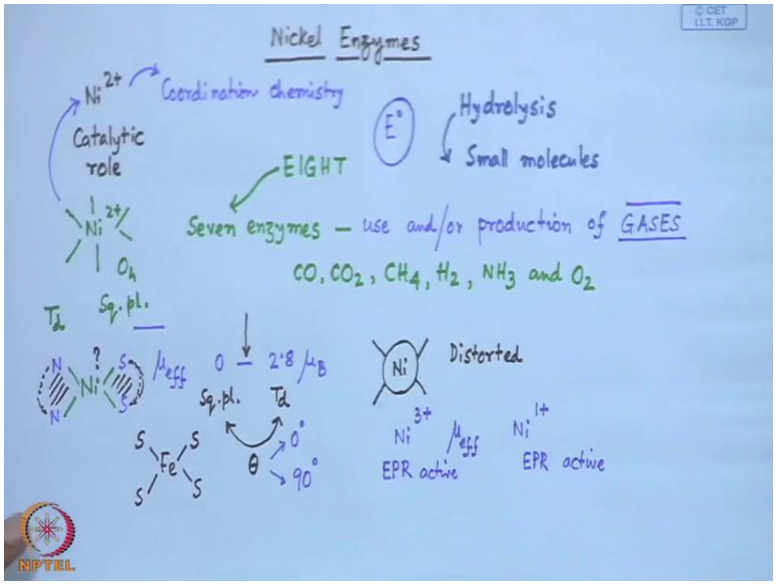

Hello, welcome to this seventeenth class. So, today we will be discussing on nickel enzymes; it is very interesting to know that how nickel place some important catalytic roles. So, there are some historical importance for these also for simple hydrolytic reaction; that means, some hydrolysis reaction and which is well known that nickel center can interact with some of the small molecules. And that ultimately gave us some idea that out of EIGHT known enzymes we have and seven out of those eight; this seven enzymes basically can interact with small molecules like carbon monoxide, carbon dioxide, methane, H 2, ammonia and O 2. So, there is a big list that all these small molecules can basically co and interact at the nickel catalytic site; and basically they are the products some time use and or production of these. So, these can be function as very good small ligands and apart from that they are all gases which is very important.

So, ultimately, what we talk about that this nickel and knowing a very good information about the nickel related to its co-ordination chemistry. Most of the time explaining all these reactions we take the help of co ordination chemistry related to nickel 2 plus. And at the same time when we talk about nickel two plus, and we know that the most

important co-ordinations side it can give it to you is a octahedral coordination side. And in some cases is some forced condition, it can also give some geometry which can be tetrahedral as well as square planer.

So, when you move from one particular geometry to the other, we get less number of ligands and some of these legends basically are bound to these particular center, and if we go for this tetrahedral and square planer geometry; suppose, if you have nickel and in a geometry like these which are bound to two nitrogen and two sulfur geometry environment. So, initially to know these environment in the leaving organization in these metal enzymes what people can find out this corresponding effective magnetic movement? So, that effective magnetic movement in a four coordinated system always gives us some idea whether you have a square planer geometry or tetrahedral geometry, because you know that it can move from 0 to 2.84. So, you have two electron paramenitism for a tetrahedral geometry and a time ending environment in square planer environment. So, if we can have some range of this say 0 to 2.8 mu b, at one end we can have this corresponding one is square planer and in other end is tetrahedral, but most of these sides related to that one particular oxidation state.

What? Right now we are thinking is as a 2 plus; that means, the nickel in plus 2 oxidation state; the most common one. So, if we get something, but if your enzyme in a particular environment because all these particular donor groups which is coming from the different polypeptide chain, the nickel sitting inside is highly distorted. So, if you have a distorted environment which is a distorted one and we get some how a magnetic movement in between. So, that gives us an immediate clue that if we have some binding pattern; that means, if we just simply co relate to our central like singular iron rubidoxin; singular iron rubidoxin was you have S S S S.

So, but here it is N 2 S 2. So, if the numbering scheme is such that these are short spanning one and these are also short spanning or the rest; that means, ns; that means, the number sequence in the long polypeptide chain; that means, if it is 88 it is 92 or 96; similarly, if it is 32, it is 36. So, this particular part we can consider as a part from the bidanted ligation.

So, if this one particular plain and this two particular plains if they are co-planer; that means, they are in the same plain, we get a square planer arrangement and if they are

perpendicular to each other, we get a tetrahedral geometry. So, these two; that means, how you can co-relate these two geometries; we can introduce some parameter which is theta. So, when theta is equal to 0 degree, we get one arrangement and when theta is ninety degree, we get another and in between we get. So, depending upon its magnetic movement suppose, if you are getting this magnetic movement is one point four and one point six you think that is there four co ordinate geometry and it is intermediate between square planer and tetrahedral geometry.

So, that gives some immediate through how the environment is there and how we can identify the nickel within the enzyme system, but apart from that if it goes because the nickel depending upon your typical corresponding potential the E 0 value, it can go to 3 plus or it can reduce to 1 plus; whether some redox catalogues is operating or not. And like your nickel in bivalent system, this try valent nickel and mono valent nickel both are paramagnetic also. So, you get corresponding magnetic movement for identifying these and also these two nickel two is not, but nickel three plus and nickel one plus would be EPR active. So, these are the basic requirement or the basic information what you can have to identify nickel in a particular center.

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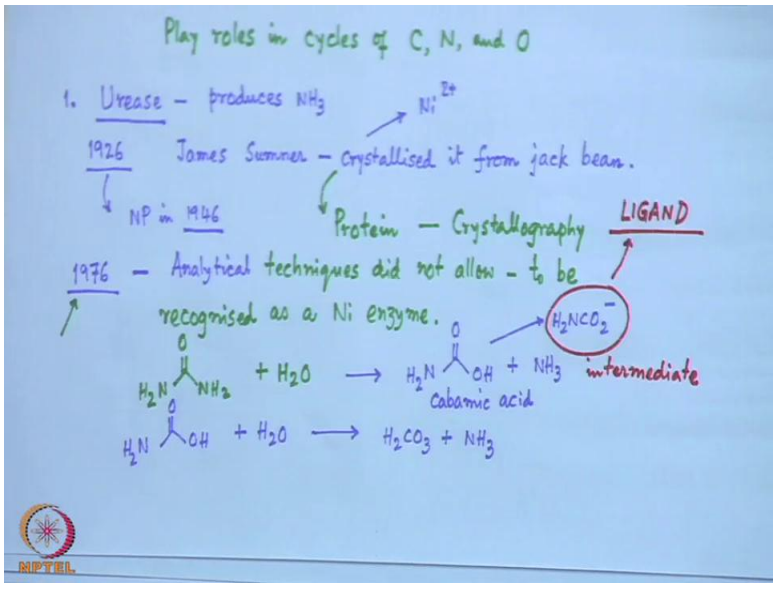

So, handling all these gases hot basically contribute to the system is that they definitely play some important roles in the cycles of carbon cycle, nitrogen cycle and Oxygen cycle. So, they definitely contribute what are the nitrogen bases species you can have in

the environment; what are the carbon base species you can have in the environment and some of these oxygen based species also. So, out of all these different molecules, one is very important is urease which produces ammonia through hydrolysis. So, it is their hydrolyzed type of enzyme and it was first enzyme which has been identified long back in around nineteen twenty six; one person James Sumner initially where was working with urease and he could crystallized it from jack bean.

So, that crystallization in that particular time; that means, is 1926, but he has no clue related to its corresponding property, but the crystallization of protein. So, it is crystallize form and which is metal bearing protein at that time it was not known. So, he could crystallize some protein at that time. So, crystallization of protein was so important, because now a days we know for determining the actual structure, we should go for corresponding protein crystallography. So, discover in that period that it can crystallize something and the metal was there. So, this crystallization was definitely dependent on the presence of metal ion. Some protein and partment and if you give metal, because sometimes the apoenzymes; that means, without the metal center it is very difficult to crystallize. So, all the metal ion enzymes and metal proteins are easy to crystallize at some point. So, the nickel bearing thing which can be easily crystallize and it basically giving something which we can go for the corresponding crystallographic pattern. So, that basically on for him the noble prize in 1946 you see the time scale; after 20 years he got the noble prize not only for doing anything, but only for crystallization of the protein, but it took 7 years until 1976; because at that time during the period of 1926 of 46, the analytical techniques were not so good such that you can examine or you can characterize this particular urease as a nickel enzymes. So, that was our limitation.

So, analytical techniques did not allow - to be recognized this particular species as a nickel enzyme till nineteen seventy six, but the reactions were known; that means, what it is doing? It is doing nothing but a simple hydrolysis of urea molecule producing one particular compound; one end of the N H 2 to group is getting hydrolyzed producing one ammonia molecule, producing a carbonic acid.

As you know that ammonia the different I means and all very easily we get the crabmeats and when you react with some ammonia oramin type of molecule with carbon disulfide, and we get also the corresponding tyanelo gas ditthio carbamates; jantids are also like that when you go for those reactions on alcohols; so jantids ditthio carbamate

and the carbamic acid. So, in the second step another molecule of urea is coming out giving instead of carbon dioxide; that means, depending upon that ph of the medium you get the carbonic acid and N H 3. So, this particular one can be when presence of ammonia or some basic condition you can consider it as carbamate and iron also; so it is the intermediate.

So, identification of these also was very crucial that this particular species either the carbamic acid or the carbamate as intermediate, because whenever we are talking all these gases and all these molecules involving this carbon nitrogen and oxygen in the typical cycle, and the metal center we always try to understand something that whether this sort of species in nickel environment or in some medal environment whether they can function as a ligand or not. Because all these transformations and all these reactions we are just talking something where you have the metal center and the ligand center. So, those two are interacting. So, this is not a very big molecule is also very small molecule having a molecular weight of say around 5, 90, 000.

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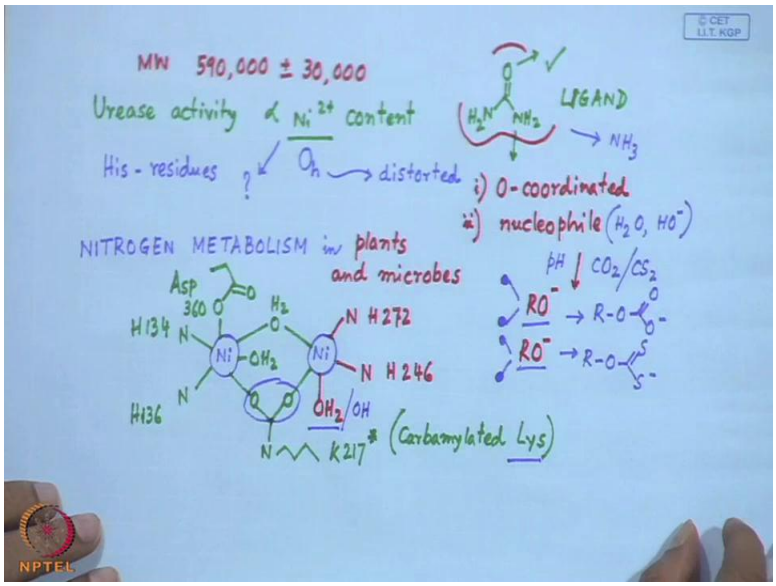

So, it had a molecular weight of 590, 000 and sometimes it varies with another small unit of 30,000, and this activity; that means, urease activity is dependent on the presence of ni; that means, is activity is propositional to Ni 2 plus content. So, it is not related to some organ catalysis that some organic molecule and some lose acid interactions are taking place and you get the catalytic activity, but here definitely you need the presence

of Ni 2 plus. So, is a metal center catalysis and a corresponding system what is present there that what are the nickel environments? So, nickel environments are octahedral and we have that slowly people have identified that you have.

So, many histedene residues and those histamine residues are attached to the nickel center giving an octahedral geometry which is highly distorted. So, you have octahedral nickel environment and around which some position is there such that the substrate, because we are looking for some hydrolysis reaction and you have nitrogen as well as oxygen; so this is your urea. So, if we think of that this can function as a ligand; so it can be interacting with the metal center though oxygen lone pair or nitrogen lone pair. So, if this particular case will see that your medal center is pound with oxygen, this environment is also facilitating that this particularly there is some pocket.

So, pocket whether that particular co ordination pocket will allow this particular end or that ends. So, this particular end is a small end and that end; that means, this carbonyl oxygen can go and directly attach to the nickel center. So, what we have once this is reacting as a ligand we get something which we can consider as O-coordinated ligand not n-coordinated. So, urea would be O-coordinated and this particular want the nickel center will show some luicacdic behavior; luiacdic behavior will be shown by nickel and basically it will drag the corresponding electron density from this oxygen, and the corresponding attack for the nucleophile will lead to the hydrolysis reaction. So, this the first step is that you have O-coordinated urea and the second step should be the attack of the nucleophile; either it is the water molecule or the hydroxide ion.

So, when we talk about that we are looking for something where your urea is consume and we are producing ammonia. So, definitely we are talking something which is directly related to the plants nitrogen metabolism. So, it would be related to our very important reaction for nitrogen metabolism in all cases; in plants as well as in microbes. So, this information's will not tell us that how many nickel centers are there within the system, but we should have some arrangement that one particular center; that means, the same nickel center can function to bound the urea molecule as well as the nucleophile; if these two nucleophile say water or hydroxide ion. So, water and hydroxide ion either they are coming from the reaction medium or it can also be bound to the metal center, but the most efficient mechanism which we can have is that the presence of two nickel center.

That means here we are talking about a binuclear system in case of iron and all other cases people have now identify that the most effective arrangement for this reactions. So, one will be responsible for substrate binding and another will be the responsible for the nucleophile binding. So, the water and hydroxide group bound to this second nickel will attach the substrate which is bound to the first nickel. So, the environment what have identified now from the structure also exes structure that these are water molecules then we have nitrogen from histidin 272 and another nitrogen from another histidin which is 246; and on the left we have one water. So, we have two water molecules attach to both the nickel center as monodentate ligands. So, that is why people are initially confused that whether the same water molecule will attack the corresponding urea for the hydrolysis reaction and also an unique bridging ligand water; two more nitrogen histidin which is one thirty six; this is another nitrogen histidin which is histidin one thirty four and this one is one oxygen from carboxy end which is as parted 360.

So, all together we have taken 1 2 3 4 5 codes inside around this nickel and four codes inside on the other, and to support this di nuclear entities, this is the modern tent coordination of single water molecule and this water bridging is also very weak if it is not symmetry one also, because this is the very long distant; this is what are the neutral one not a hydroxide one or oxide one. So, we need some other support from this side which is the car boxy bridging and which is not a pure carboxylic group, but is carbymilated lysine. So, which is numbering is K two one seven and one star, because this is derived one; you have the lysine and that lysine group you have N H 2 function, and N H 2 function is reacting with carbon dioxide giving that carbonylated lysine. So, these are some unique transformations in all these living system that if you have V N H 2 N or some time, if you have in synthetic molecules what you find that if you have R O minus that any alkoxide group.

So, these alkoxide group can reach two medal centers like hydroxide an oxide these also can reach two medal center, but if in between you have certain arrangement that if your ph is suitable and in a carbon dioxide environment or if you can react with carbon disulphide the bridging groups like R O minus and R O minus can be transformed to the corresponding alkyl carbonate R O C O OO minus; this also zentet Z double bond S minus. So, we were just thinking or we are talking that these simple groups can bound two medal centers; one medal center here and another medal center was another medal

center, but sometime presence of this carbon dioxide or any other group this bridging groups can be transform. So, if you have a free a minus like a lysine with N H 2 function and that N H 2 function with the use of carbon dioxide, it can be converted to a corresponding carbonic acid type of arrangement and that carbonic acid type of arrangement.

Basically this is the strong binding compare to your water binding and these two can keep the nickels this a separation also unique for binding of your urea molecule, and the corresponding attack from either these water molecule or its corresponding deprotonated one. It can go for immediate deprotonation also to give you the hydroxide to bound nickel one and it can attack the corresponding binding of the urea molecule to this center. So, this particular hydrolysis reaction though it looks very simple, but it has some kinetic barrier.

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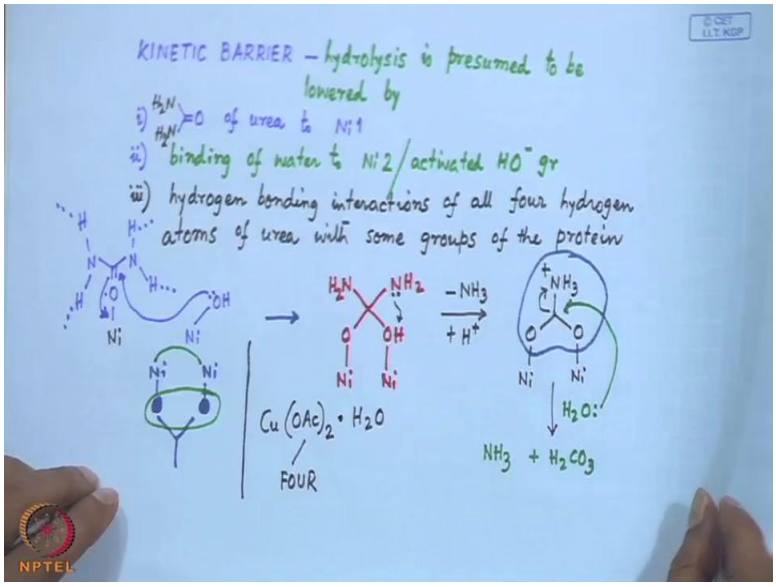

So, if you have a huge kinetic barrier for any reaction basically that can be lower by all these coordination and other thing. So, barrier of this kinetic reaction for hydrolysis is presumed to be lowered by the first one which we just talked right now, that coordination of this carbonyl group of urea to one nickel say nickel one. So, binding of water to the second nickel; that means, when it is bond to water it can undergo depot nation and you can have some activated means is nickel of activity is increased; you have activated O H minus group in your hand for the reaction.

And lastly which is also very important when you have the corresponding substrate and this substrate we all know that the urea's oxide is very interesting to understand the hydrogen bonding interactions. After all these coordination you still have some hydrogen bonding interactions, because this O group is bond to the nickel center, but if you have the N H 2 here and which are away from the medal side, and these two nitrogen this N H function can be grouped hydrogen bonding donates. So, you have hydrogen bonding interactions of all four hydrogen atoms of urea with some available electrophile groups of the protein. So, protein will be responsible for suitable positioning of the entire urea molecule not only two the metal coordination, but hydrogen bonding interaction with these N H 2 functions basically grabbed the entire molecule close to that of your to nickel centers; some groups of the protein.

What we have seen in case of adjoining also which is the cationic one, but still suitable positioning is allowed close to the metal side. So, one nickel will be responsible for binding your carbonyl function and these hydrogen's will show hydrogen bonding interactions with some other groups of the protein envelope, and the second nickel center will have the O H group.

So, this will attack this carbonyl function, because we just go for electron push to the medal center. And as a result we get some intermediate, because this intermediate when this O H is attack this carbon becomes tetrahedral. So, we have O and this is attacked O H. So, this one to first nickel and this one already bond to the second nickel, and this non pairs are available. So, it can take up this as a proton removing one ammonia molecule and this particular one in for if the system also gives one proton. So, this can be either N H 2 or N H 3 plus if you take this proton. So, in one particular site you have the carbamylated lysine. So, you also see something the same type of arrangement the bridging groups from this thing; that means, what we are looking for the product formation for hydrolysis of the urea. So, this ultimately is responsible for the removal of the second molecule of ammonia when your H 2 is attacking. So, at this particular point you have N H 3 plus H 2 C O 3.

So, the interesting part for this is that you have the corresponding support; that means, this carbamylated type of arrangement which was there on the other side of the nickel. So, the thing is that you have the nickel environment and you have the corresponding bridging groups on this side. So, this nickel center is there and this nickel center is there.

So, other side also similar arrangements is taking place. So, initially within a ligand system whatever groups are presence; that means, a weakly bond water molecule which is loosely bond as well as the another mordent at water molecule, all will go. So, basically the enzyme is resting on and acited bridging moiety.

So, in this particular want because the enzyme is a very big molecule and whatever thing we should think of; that means, if we just talk about some model chemistry or synthetic chemistry related to this let if we can get this particular arrangement; that means, for getting the arrangement for copper acetate; all we know this are the very standard example and well known example, but still we do not know all the time we do not care all these thing. So, this copper acetate monohydrate which is basically a demand and when you have two copper center like these two nickel what you get? You get four breezing acetate groups.

So, this four acetated groups are basically responsible for the stabilization of a binuclear entity, but in the protein environment what we are talking about and what we are discussing that one you have these when sub state is coming and binding to the nickel center; as well as your niclo file is binding to the second nickel center, the entire thing within the protein envelop is supported by only one acetated type of bridging which is very difficult to get; because you may argue that why we are not getting one particular arrangement that all other ligand positions you can change starting from simple coparasited.

So, you remove one acetate first by some other donor group; you remove the second one; you remove the third one; only I will remain with the binuclear arrangements with one acetated bridging, but that is not so easy and it is also impossible, but in this protein environment that you get something where your one side is breezed by the acetated group and whatever sub state; that means, that corresponding driving force for going for this sort of sub state; that means, this bridging entity is also supporting this binuclear entity, because on the other side this bridging is forming.

So, as the intermediate after removal of the fast ammonia molecule thus before the removal of the second ammonia molecule whatever arrangement you can have; that means, in this side you have this already carbamylated bridge and the other side you have the substrate; the transform substrate is breezy. So, this particular type of

arrangement is still there and which is a stable arrangement. So, all these things will give something that you just go for the corresponding rate of transformations and the corresponding kinetic parameters are also very high in number, and we get immediate evolution of this ammonia molecule, and all these.

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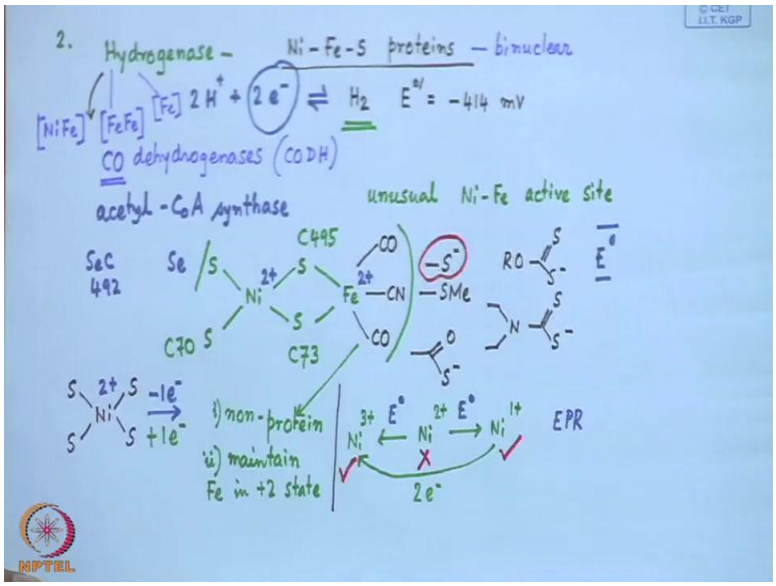

This is the first one. The second one which is a very important one for some important reactions like hydro genesis. So, once we get that urea's, the second one based on nickel. So, nickel enzyme will be useful for hydrogenose reaction. So, we are handing carbon dioxide and ammonia now we are talking about the hydrogen gas. So, all these gas has reaction and that particular reaction will be just simple; though it is a very simple reaction and in terms of our N H demand how we make this simple reaction.

So, it is a redox reaction. Now, the difference with that of urea's and we still use the nickel center. So, is a two electron redox transformation for production of hydrogen from proton and E 0 prime is minus four one four mini volt. And for this particular arrangement we not all the need nickel, but one well known system is based on iron and sulfur. So, there are several other hydro geneses. So, large number of hydro geneses are known; there are some hydro geneses which bears only iron and sulfur. So, they only iron bearing hydro geneses, but when you talk about nickel enzymes and nickel will also be present along with iron. So, it will be nickel iron sulfur proteins.

And not only they are useful for the reactions of hydrogen productions from the proton, because it is highly negative potential. So, we need some corresponding difficult reaction for the electron transfer to the proton. So, not only hydro geneses, but also this sort of molecules; that means, the nickel iron sulfur proteins are also useful for carbon monoxide dehydrogenizes. So, we will see one after another slowly and they are a variegated as CO DH. So, again after ammonia, after carbon dioxide, after hydrogen, we are talking something again another gas molecule. So, you see all these things because all these nitrogen proteins and nickel proteins, and nickel enzymes are useful in some an aerobic condition; that means, we do not need oxygen for all these transformations. So, these hydro geneses can be considered as we just within square bracket with write it as nickel iron hydrogenise there are one type which is Fe Fe hydrogenise; that means, when you write Fe Fe hydrogenise; that means, at least two metal centers you require. So, all whatever we are talking on all these proteins they are binuclear.

So, we need two metal centers and sometime only one iron center is also sufficient for hydrogenise reaction. So, one such is after hydro geneses is similar nickel iron sulfur protein is useful in carbon monoxide de hydrogenise and another very important reaction that we discuss in our next class in acetyl co enzyme a formation; acetyl it is nothing but how you make the acetyl function from carbon monoxide and carbon dioxide, because we are talking something that how we get organic molecule through these biological pathways starting from either carbon monoxide or carbon dioxide. So, acetyl function you can synthesize. So, acetyl co enzyme these are the co enzyme basically not coward. So, co enzyme a synthes; thus this is the responsible for the synthetic of acetyl co enzyme a.

So, let us first see about these hydro geneses. So, this hydro geneses are bearing nickel and iron. So, basically is a very unusual one also; unusual nickel iron active site. So, both the iron sites as well as a nickel site will be responsible for the reaction; that means, you have to have some arrangements whether you have the corresponding hydrolytic cleavage of the hydrogen atom or some holytic cleavage of these hydrogen atom; as the hydrogen a hydrogen molecule as a molecule atom and those groups basically how they are going to bond to the metal center or some other groups nearby two those metal centers.

So, First of all we have to identify the groups which are binding our two metal center; that means, nickel as well as iron and the identification of their oxidation states. So, in the resting state both the two metal centers for these is plus two on nickel and plus two on iron, and that unique bridging groups are sulfur. So, that is why it is nickel iron sulfur protein; we know all about the iron sulfur proteins. The ferredoxin the rubredoxins and all other. So, it is also a nickel iron sulfur protein, because you have this sulfur and this sulfur aim is coming from the sixteen residue with sequence numbering is 495 and 1673. So, they are quite a part. So, if you starts from here, it goes and the other side it is then ultimately coming at 495, and here the other two positions one is again system and which is systemate seventy, and other is sometime sulfur; from the systin residue than some other cases it is selenosistine; that means, selenium function is there.

So, selenosistine residue number 492; some molecules it is selenium which is very much close to sulfur. So, this particular environment; that means, this side you have nickel in a typical sulfur environment. That is why during the last twenty five year or so people are in hesitating so much about the corresponding words in chemistry of nickel in typical sulfur environment; that how many sulfur environment you can have with the sulfur is typical S minus or thaivithar sulfur or thaiyomite sulfur or any other thairidia type of sulfur. So, if you have the different sulfur groups attached to the nickel center and people just simply try for that the typical reaction and the model chemistry that how many of these in sulfur environment can go for either corresponding a typical lose of electron; that means, either it go for plus 2 to plus 3.

So, when you for oxidation; that means, whether your sulfur environment can support the oxidation of the nickel center. So, people make some different types of ligand system; we make the mononuclear compound, because initially the challenge was how you find out this particular mononuclear environment though it is a part of the bi nuclear environment system, but it is a mononuclear environment. How it is behaving if there is something which is the redox reaction? Because this is a typical redox catalysis. We have to supply the number of electrons to the proton center and if you have some proton pool; that means, each plus is there and interestingly we will find that some of these age groups can also be bound to the sixteen residues. Because one such good example for the ligation for this nickel center is the sixteen sulfur; that means, S minus.

So, your S minus can be a very good ligand. If you go for the corresponding alkyl group; that means, the S methyl function that is also a very good ligand for you and sometime you can go attach some carbonyl function, and then S minus. This is also a very good environment for sulfur; then you can have the corresponding jenthas environment; then you can have N and diethyl amine which can react with these sulfur groups to give you the dithiocarbamate functions. So, these are some sets of all these molecules what can give rise to some coordination chemistry for nickel in sulfur environment. And what people can do that for the oxidation as well as the reduction reaction you do not know what is there, because sometimes you will see in these catalytic function that you have to support all three oxidation state. So, your nickel environment is such that you can go down to nickel in the plus one oxidation state or you can go up to nickel for plus three oxidation state. So, during this catalytic turnover if you just simply go for these we got you think it clearly that in case of iron and in case of copper during that the di oxygen activation in hemocyanin; we get the reduced form. Then immediately you transfer both the two electrons to the corresponding anti bonding or baital of di oxygen molecule and it transform the di oxygen molecule from di oxygen itself to peroxide. So, those electrons are basically we are feeding to the di oxygen molecule in its anti-bonding or baital.

Now, what we are doing we are doing nothing but we are pushing those two electrons to the protons. So, basically supplying those electrons to the protons will ultimately lead to the hydrogen production. So, if we just simply go from here to there; that means, if you just can make something where you have the nickel in plus one oxidation state immediately you can go to nickel plus three oxidation state, you get two electron out of this. So, this two electron transferred is possible and those two electron transfer will be require for utilizing your reduction for the protons to hydrogen molecule. So, in this environment what people did for all these of ligands? Different types of ligands sometime with the combination of nitrogen toner as well as oxygen toner. So, you make the synthetic molecule and you determine the corresponding E 0 values by simple technique like cyclic voltammetry.

So, cyclic voltammetry will tell you in the next step what are the corresponding E 0 values how easily you can go from a nickel two plus compound, a bivalent nickel compound to a monovalent nickel compound or a trivalent nickel compound. So, you determine these E 0 values. So, the knowledge of these E 0 values will tell you how

easily you can go from one oxidation state to another? How easily the other two oxidation states are accessible from bivalent nickel? And also some spectroscopic technique like electron paramagnet resonance; EPR is useful for identify the formation of nickel in plus one oxidation state or nickel in plus three oxidation state, because this is not EPR active, but this is EPR active and the trivalent state is also EPR active.

So, all these chemistry little bit you can study also that the corresponding cords in chemistry in all sulfur nickel environment which is very important and from all sulfur nickel environment you can go for a binuclear entity also, because if you have this s minus thus we charge is more on these groups. So, if you have the sixteen sulfur over here and it is neutralizing the nickel. So, you have 1 2 3 4, four negative charges are there which are not sufficient for going for this corresponding charge neutralization on the nickel two plus. So, either it can for a oxidize form in the plus three where the overall charge on the complex would be less. If you go for oxidize form or this sulfur can be used for bridging the second metal center; that means, this particular group this ligand groups will have some tendency to breech the second medal center.

So, why these two sulfur groups the sixteen sulfur groups are different compare to this sulfur which is in the terminal position. So, iron site you get. So, these two sixteen sulfur residues are attached to this particular nickel side and you have see other groups which are attached to your iron site. So, these are now very unique which is not known earlier in a living system that these are CN, CO and CO function. So, these are typically non-protein ligands. So, whatever in organic chemistry you are doing in the laboratory we see in the living organism also. So, these three ligands CO CN'S and CN they are therefore, non-protein in nature. So, they are non-protein ligand which is a very unique system for this sort of molecules. So, whatever we are talking is the protein molecules, but the ligands are of different origin.

So, they are carbon monoxide, they are CN and another carbon monoxide. So, they are non-protein ligands and these ligands all we know from the typical coarsen and chemistry knowledge that they are responsible for stabilizing the metal center in low oxidation state. So, they are all pyosi ligands we all know. So, this three ligands basically restrict the iron center in plus two oxidation state. So, they basically maintain the iron site in plus 2 oxidation state. So, if you know the basic structure from in this fashion that is identification of all these positions around iron will immediately tell you that where

from you basically get the redox activity; whether this corresponding redox activity you know both the medal centers are redox active; whether you get the corresponding redox activity from the iron center or the corresponding redox activity from the nickel center.

So, once you stabilize or just block this corresponding a center in the plus two oxidation state; that means, this center cannot go to plus three oxidation state. So, definitely you whatever redox activity related to the reduction of the protons we should get from the nickel center. So, nickel center will definitely supply the electrons for the reduction of all these protons; it is not the iron center. So, that we will see in our next class that how the catalytic cycle is operating in that way.

Thank you.