Bioinorganic Chemistry
Prof. Debashis Ray
Department of Chemistry
Indian Institute of Technology, Kharagpur

Lecture - 2
Iron storage and Transport – I

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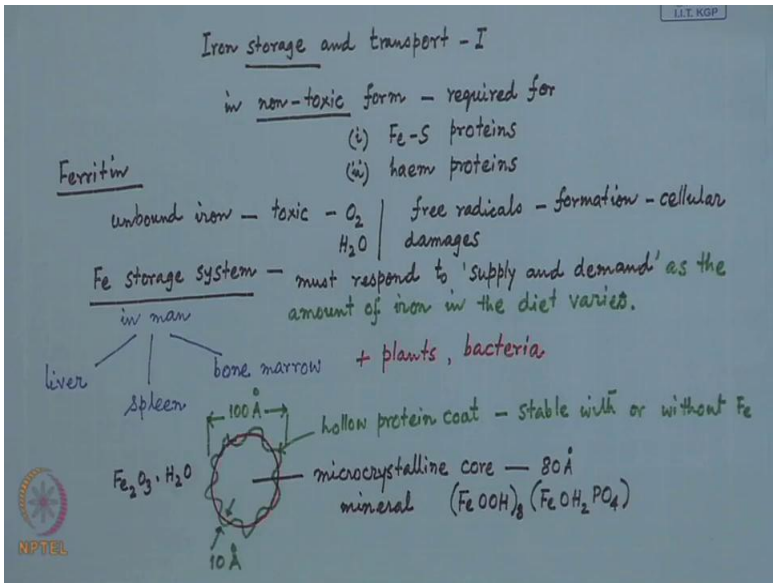

Hello, today we will just as a continuation, start on iron storage and transport - I. In this section we will just see how storage of iron is important. Particularly in some of other living systems were in non-toxic form, it is required for we all know now that most important iron containing biomolecules. Because why we need this iron for the synthesis of several iron containing biomolecules what we have seen starting from hemoglobin and all.

So, the first one we can just right in this form that you can also utilize this for iron sulfur proteins synthesis of iron sulfur proteins where we get the regular supply of this iron from a storage system. So, how this storage system we will get that we will see in this class. Also we can utilize it for the synthesis of different hem proteins such as your myoglobin, your hemoglobin, your cytochrome c and all these things. So, all these molecules where we need the iron for the synthesis of a particular iron containing biomolecule, we want to store this particular iron first.

So, one such important biomolecule for the storage is ferritin. So, this ferritin molecule we will just see that how this iron can be stored and in most of the cases this non-toxic form of this iron is important, because in most of the cases the unbound iron; what is that? Unbound iron means were we do not have any chelating group which is binding your iron center; that means the free iron, what we get actually in case of a solution of iron in a typical aqua solution or acidic solution of slightly basic solution. So, this unbound iron in most of the cases particularly in the biological system is basically toxic in nature because of the presence of O 2 and H 2 in the system where O 2 can take off one electron, it can oxidize the iron center and H 2 O can control the corresponding pH values.

So, in all these cases the unbound iron which is free in nature which is not bound to the ligand system, they are mostly responsible for the generation of free radicals. So, we get different free radicals and these free radical formations are therefore related to the accumulation of large amount of iron in to the lathing organisms. So, this formation of these free radicals is also important to know that because these particular free radicals will be responsible for different types of cellular damages. So, we want to stop that cellular damage. So, we want to stop the formation of these free radicals and we want to get the corresponding bound form of this iron. So, this bound form of iron how we get that we want to have a particular iron storage system.

So, this is therefore our iron storage system which is our ferritin molecule and why we want to store that, when we get this particular iron from our food material or any other supply source and that immediately required for the synthesis of, say, iron sulfur proteins or hem proteins. So, we do not know that how much iron we require for the synthesis of these proteins molecules and how much iron we have in our system. So, there will be a continuous demand and supply process. So, this iron storage system such as the ferritin molecule therefore should respond or must respond to supply and demand as the amount of iron in the diet varies; that means if we have some excess amount of iron which we are consuming from our food material in to our system, then the excess iron can be stored.

But if we have the less amount of iron what is required for the synthesis of these protein molecules what we have to do? We have to supply this particular iron from your store house. So, these ferritin molecules are therefore important to give us this corresponding

supply and the demand as and when we use these particular iron sulfur protein molecules to be synthesized in the system. So, this particular iron storage system what we have, particularly in our human body, they are stored in three different places; one is liver, another is spleen, and third one is most important place you know all where the synthesis of hemoglobin molecule is taking place; that is your bone marrow.

So, we all should know that where the concentration of these ferritin molecules. We are talking in one hand the iron concentration in this ferritin molecules, at the same time how much ferritin is stored in all these important places like liver, spleen and bone marrow and not only in the human system, it is also present in plants as well as in bacteria. So, this particular thing; that means we should take the help of the protein molecules and protein molecules will selectively collect this iron centers and they will be stored in typical process where large amount of iron centers are stored inside the ferritin molecule.

So, what will have? We will have a spherical system and this spherical unit will have a typical hollow protein coat and this hollow protein coat will give you a spherical structure of about 100 angstrom diameter and the thickness of this protein coat is also known which is 10 angstrom. So, this hollow protein coat is very important because it plays an important role, because this particular hollow protein coat is all the time it is stable with or without the presence of iron.

So, you have a protein coat and if you get this iron centers and this iron centers if they are allowed to enter within the coat, we get the corresponding assembly of this iron anions in one particular form and as a result we get inside this particular coat, a microcrystalline mineral coat which is microcrystalline in nature; that means you can use extra diffraction to identify the composition as well as nature and the morphology of this particular microcrystalline coat. So, this microcrystalline coat typically has a size of 80 angstroms and the composition of this particular mineral coat can be determined as a hydrated form of ferric oxide.

So what we are getting from the system is the ferrous or ferric ion which is ultimately oxidized and depending upon the availability of the water molecule, it can undergo the corresponding deprotonation reaction and that deprotonation give slowly one after another iron center and this iron centers will ultimately give you something which is very similar to that of our iron ore or mineral, hematite or magnetite. So, in the biological

system also the same process is followed where from the basic ferric iron or the ferrous ion we get a coat structure. So, which has a very similarity with that of your mineral and composition can also be determine from analysis that we can write down the formula as Fe OOH 8 and Fe OH 2 PO 4; that means not only iron you have the presence of oxide anions as well as the hydroxide anions as well as the phosphate groups are there and this phosphate groups are basically terminating the corresponding chain.

So, as it grows from iron oxo, hydroxo species we get a chain like structure and that chain like structure ultimately giving rise to a two dimensional network and that two dimensional network is ultimately attached to your protein sib or protein envelop. So, you have the hollow protein coat and the protein you can identify different lead; what is the nature of that protein and how we can find out the corresponding protein structure for that coat and what is there inside as a microcrystalline coat. So, this composition is determined and that composition is similar to that of you have phosphate, you have oxo, you have hydroxo groups and you have the iron centers.

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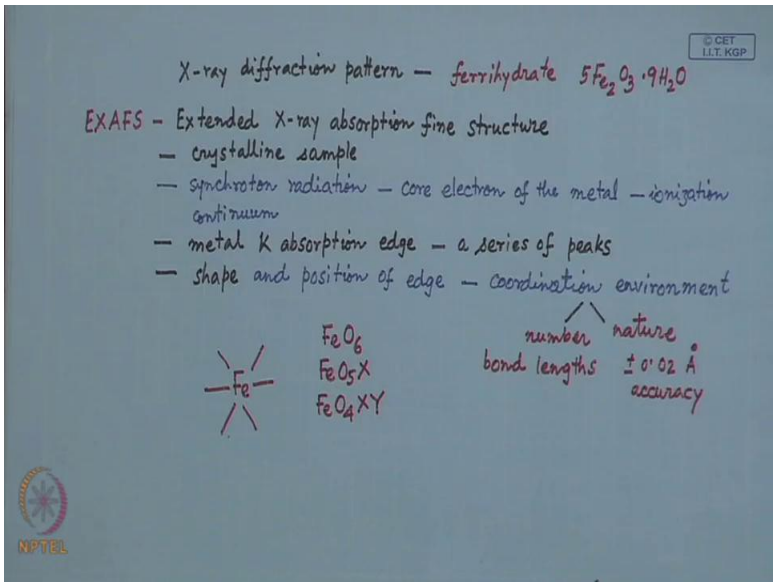

So, x-ray diffraction pattern very well it can utilized for the identification of the typical composition of this ferritin molecule, and if we compare this x-ray diffraction pattern which is the powder x-ray diffraction pattern not any kind of single crystal x-ray structure determination. So, for a powder x-ray diffraction pattern of the ferritin code what you have in your hand, you determine the corresponding x-ray diffraction in the

powder form and you compare with the know iron minerals what we can have in our hand and that basically gives us that which is similar to that of your ferrihydrate molecule which is known to us which is 5 Fe2 O 3 9 H 2 O. So, basically its structure as well as the morphology is similar to that of your mineral ferrihydrate.

So, one particular technique what is your x-ray diffraction as well as we can take the corresponding IR spectra to determine that iron oxygen stretching frequency or the iron hydroxide stretching frequency and the phosphate stretching frequency, the bond stretching frequency can be found out. So, what are the basic ingredients what are present in the ferritin molecule can be found out. Next another physical technique which is very much useful for the determination of the nature of the ferritin code is EXAFS which is an EXAFS absorption technique extended x-ray absorption fine structure. So, the abbreviated form of extended x-ray absorption fine structure. So, one typical advantage for this particular process of absorption spectra is that it does not require any crystalline sample.

So, crystalline sample is not required for single crystal x-ray structure determination; we always need single crystals for the determination, but in this particular case we do not need any kind of that sample. So, sample preparation is easier because most of these protein samples sometimes very difficult to crystallize it. So, biological samples most of the time it has large amount of water molecules. So, is very difficult to crystallize those materials. So, we use synchrotron radiation to excite the core electron of the iron center or any other metal center which is a very useful technique for determining the nature of this corresponding biomolecule.

So, any core electron of any metal center can be excited to its ionization continuum. Then what the next thing we determine is the metal K absorption. Edge is determined and what we get a series of peaks and those series of peaks like any other x-ray diffraction and powder x-ray diffraction probably you know all these as well as the IR spectrum. So, all these peaks can then be corrected as for their shape and position which basically gives us the required information for the coordination environment around the metal center. So, it gives us the coordination environment which we cannot get from powder x-ray diffraction or FT IR spectrum.

So, this number and nature of this donor atom. So, once we get the corresponding coordination environment we should have corresponding information. So, what is present in the immediately first coordination environment of, say, your iron center? So if the ferritin is present there, so what are the donor atoms which is immediately present and which is surrounding this iron center. So, where your iron is your 4 coordinated one or 5 coordinated 1 or 6 coordinated one and depending upon the available number of donor atom around the iron, you should know the corresponding geometry and that particular geometrical information will at the same time will give you the corresponding rough estimation of the corresponding metal donor atom distance.

If it is surrounded only by oxygen, so what is your iron oxygen distance? So, a rough estimate of your iron oxygen distance and the number of oxygen atoms which are surrounding your iron center can be very easily found out using this particular technique. So, is a very simple technique and there is no need to give you a corresponding information related to only the crystalline samples what we get by determining the x-ray structures So, for determining the corresponding coordination environment. So, you should have some information about the number and nature of the atoms; that means the donor atoms. So, at the same time that also gives us the bond lengths and bond lengths determination is pretty accurate with a deviation from plus minus 0.02 angstroms accurate.

So the problem what we can have is you have the iron center. So, from this EXAFS data we can establish that weather you have a environment of Fe O 6 only or an environment which is Fe O 5 X or environment which is Fe O 4 X and Y; what are these X and Y? Sometimes we will find that some of the donor atom which cannot be identified precisely. So, that can be chloride, that can be phosphorus or that can be any other donor atom. So, those atoms we can match the corresponding bond distances; that means related to your iron oxygen bond, if we can determine by this EXAFS technique the corresponding bond lengths Fe X and Fe Y, then we can find out that this particular center can have a different geometry or different orientation of all these groups.

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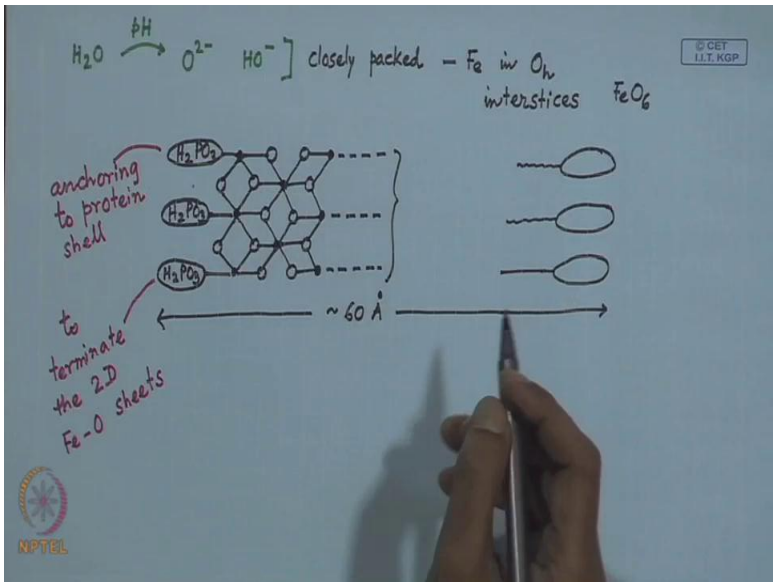

So, this particular center when we get within the matrix of some groups which are nothing but our O 2 minus and HO minus. These groups are derived from water molecules attached to the iron center and definitely it is dependent on the pH of the medium. So, pH of the medium will tell you whether you can have sufficient supply of hydroxide groups or sufficient supply of these oxide groups which can bind your iron center. So, immediately when you have the corresponding hexaaquairon center after deprotonation very first step of this is the immediate dimerization of the system. So, dimerization will take place for hydroxide bridging. So, Fe OH Fe; so that Fe OH Fe unit where your O H group is bridging to iron centers, so that can be very easily identified in your excess data also; that means you have Fe O H distance which is different from your Fe OH 2 distance and Fe O distance of oxido group.

At the same times you can have your Fe O Fe bond angle how they are forming. So, is a three dimensional structure. So, that particular bond distance as well as the bond angle will tell you what is your nature of the corresponding group, which is directly attaching to your iron center. So these two groups O 2 minus and HO minus, they are then closely packed. So, they are basically the regular sphere what we know, the structure of the different mineral like your hematite and magnetite. So, these are closely packed and iron is present their in octahedral interstices. So, most of the cases what we will find is that this iron is in a Fe O 6 environment. So, what is the role of our H 2 PO 3 units which is

basically attached to the iron center? These hollow spheres are oxides and hydroxides and solid spheres are iron centers.

So, this network will form immediately. So, this particular 2 D chain will continue. So, one half will be at the left hand side, this will go the other half there. So, this will continue from left to right. So, on the right hand side what we will have? We will have the same continuation of this and these terminating groups. So, the dihydrogen phosphate groups basically will terminate these 2D chains and that entered chain length is also known to us which are of 60 angstrom length. So, these phosphate groups they are basically functioned to attach to the protein cell. So, they are basically anchoring to protein cell.

So, what we have? The protein structure, then the phosphate groups is coming out from the protein cell and you have the inside. So, depending upon the amount of iron which is being injected within the hollow cavity of the protein cell you have a continuous growth of this. So, basically it is a typical assembly process; assembly of the ions, iron centers in presence of the oxides and hydroxides you have a huge amount of this particular growth. So, how long this growth will continue; that will be dependent on the cavity size of this particular hollow sphere. So, when it is reaching to this phosphate ends it basically stops the growth there.

So, within the code you have a nucleating point; that means the first iron center which is coming inside and which is attaching to the second one, you get a dimeric form, then you get a corresponding trimeric form. So, that is why I showed you in the last class that if you have water, then that water after deprotonation giving you hydroxide function. So, that hydroxide group is attaching two iron centers, but that OH function can function as a mutually bridging unit and when it is functioning as mutually bridging unit it can bind three metal centers. So here also like this, these are your hydroxido function or the oxido function which is attaching to one to three iron centers. So, this particular form. So, it is continuing in the two-dimension as well as it will continue into the three dimension.

So, these phosphate groups. So, not only it is anchoring to the protein cell, but it also used to terminate the 2D structure; that means 2D Fe O sheets. So, you get a typical sheet structure and that sheet structure is being terminated by the phosphate ends and we now know that how the typical structure is generated because you have the iron at this point

and you have the hydroxide or the oxide function. So, both the combination of these two will generate the corresponding 2D network and that 2D network when we get that particular form the protein structure.

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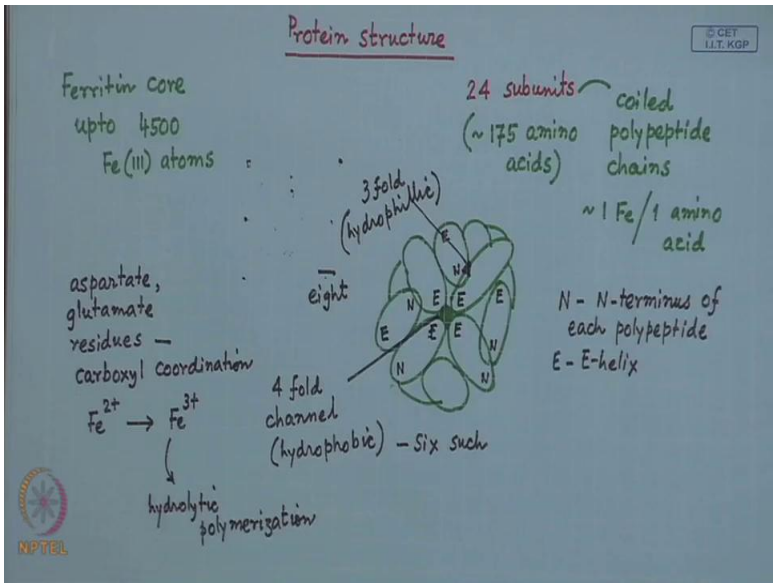

So, the analysis of the protein cell gives us some idea that it has 24 subunits. So, a long polypeptide chain is getting coiled and giving a one particular polypeptide chain. So, these subunits are nothing but coiled polypeptide chains and the length of one particular subunit is also known and which is forming from 175 amino acids. So, in the ferritin code not only the composition but also we can determine the percentage of iron also and from that determination we can also find out the number of iron atoms present within the code and that is also a very weak number. So, it can accommodate basically up to 4500 iron III atoms and you see now you have 24 subunits which has a typical three dimensional structure that we will see and that three dimensional structure we next accommodate the 4500 iron atoms.

So, basically what we get? We get that about one iron is there par one amino acid. This is basically the ratio of amino acid as well as the corresponding iron center. So, you see the large amount of amino acid which has been coiled to give a polypeptide chain and those chains are required to get a corresponding protein cell and those cells will accommodate the huge number of iron centers within the polypeptide chains. So, no we will see how these 24 subunits are located in a three-dimensional structure. So, it is even number

arrangement of all these groups. So, basically it has some cemetery and this cemeterical arrangement is also very nice to know. So, this is one particular coiling. So, coiled polypeptide chain. So, you have a long polypeptide chain which is getting coiled and basically a long oval shape structure it is generating.

So, 24 substructures you can have. So, this is one and then you can have another on this side, then you can have another on this side. So basically how I am placing all these things because you have a fourfold cemetery. So, you have in a square arrangement we are placing all these groups and other ones is also overlapping; some amount of overlap is there. So, you have a sphere like arrangements of 24 subunits in three dimension. So, arrangement of these 24 sub units is very easy to identify because you know all the corresponding geometry, suppose you have a typical octahedral. So, in one particular trigonal phase how many phases you can have? Eight; so 8 trigonal phases you can have in octahedral geometry. So, eight; so on one particular trigonal phase if you can accommodate such subunits three times; that means 3 into 8 will give you a 24, number is 24.

So, arrangements of these are typically a spherical type of thing because an octahedral geometry also has a spherical cemetery. So, the arrangement of these will give you immediately what we are generating and some of these points are leveled as E and N. The coiling of these depending upon the amino acid residues present in all these coiled chains. This is N, this is E, this is N, this is E, this is E, and this is N. So, at this particular point it is a fourfold cemetery. So, at these fourfold cemetery you see all these four E ends are attaching there. So, N ends are N terminus of each polypeptide. We have 24 such polypeptide chains and E is a typical helical structure of this polypeptide chain we considered as E as the E-helix part. So, this fourfold channel which is N terminus of each polypeptide, so these each part is basically we get from here the fourfold channel.

Why you are talking about this channel? Because how you have a protein envelop a spherical arrangements of the protein envelop and how the iron centers are entering there because you have to put iron center. So, there must be some channel and through those channels iron will entire into the ferritin code and form the corresponding mineral like structure. So, you have a fourfold channel and that fourfold channel is basically a hydrophobic in nature. So, this particular hydrophobic part of this center will not allow you to enter the corresponding iron ions; so for that we need some hydrophilic channel.

So, you have fourfold channel and this fourfold channel are there; total numbers are six like your octahedral arrangements, you see your c 4 cemetery of octahedral.

So, you have X Y and Z, but X is not allowing to the same thing because one X is different positive direction and negative direction; they are two different. So, 3 into 2, so altogether you have six such channels which is similar to that of a typical octahedral arrangement. Then on the side of these, basically somewhere here, you have a threefold channel which is hydrophilic in nature. So, how many would be three fold, 8 such threefold channel will be there. So, these threefold channel are made up of residues amino acid residues like aspartate and glutamate residues; that means we have carboxyl coordination.

So, that carboxyl coordination will give us the charged coordination and those charged coordination will be useful for our iron entry. So, iron is there. So, which is if they are coming as Fe2 plus will be immediately oxidized by some other enzymes to Fe 3 plus and they are entering through this threefold hydrophilic channel and those threefold hydrophilic channel will then fulfill and the inside code of the ferritin molecule will be filled through this iron, hydroxido and oxido species and the process what we can consider is as their hydrolytic polymerization. So, through these channels this irons ions are entering within the ferritin code.

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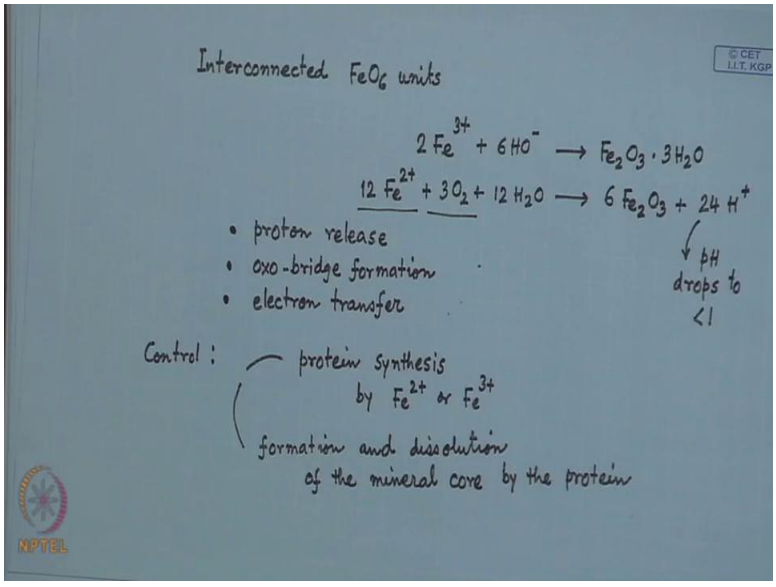

And those codes are identified again what we learnt through that technique EXAFS that they are basically providing some interconnected Fe O 6; they are all interconnected. So, individual Fe O 6 unit what we are getting there, they are all interconnected and during the early stage of code formation the chemical reaction what is taking place there are very simple reactions of Fe 3 plus with 6 hydroxide groups giving us Fe2 O 3 3 H 2 O or at the early stage of code formation, we can also utilize the ferrous ion and the ferrous ion can be oxidized by the dioxygen of air and in presence of water molecules; sufficient water molecules are there to give us again the ferric oxide with liberation of large number of protons which is very important.

These large number of proton liberation from this reactions basically changes the pH of the medium because the pH controls some important role for iron solubilization. We all know that how we face difficulty in solubilizing the ferric chloride in water. So, inside the cell and outside the cell you have a tremendous different in concentration. Sometime concentration drop down to 10 to the power minus 18 molar compared to a concentration of 10 2 the power minus 3 4 or 5 molar of iron concentration depending upon your pH. So, within the cell your pH value due to the libration of this number of proton, the pH sometimes drops to less than 1. The medium is so acidic in nature that this particular proton can control the corresponding formation of the corresponding ferritin code.

So, basically during this process we should consider the proton release during the formation because we do not have any other externally added base which is taking care of this base that corresponding protons what is getting released from the reactions. Next is OXO-bridge formation and at the same time like the oxidation of ferrous ion by atmospheric oxygen we have to consider the corresponding electron transfer reaction. So, when you study the iron transport this particular class we are just studying the iron storage. So, during the transport, so when you we have the hexaaquairon we know what is the difficulty in oxidizing that particular center from ferrous to ferric and if we take the help of some of the ligands which are bound to your iron center and which can modulate the corresponding E zero values.

So, during binding of these ligands we can change the corresponding potential. So, depending upon the available oxidizing agent. So, some oxidizing enzymes are there which are basically oxidizing this particular ferrous ion by this dioxygen molecule. So, all these together can give us some information how this ferritin codes are forming and

side by side we can just match the synthesis of the typical iron oxo-code in the laboratory such that how this iron is stored inside the ferritin code and how we can control the formation. So, iron is stored in the protein ferritin and the protein code is a hollow sphere and two things are there at this concluding part that is which can control. So, one is your protein synthesis and that protein synthesis will be controlled by Fe2 plus or Fe 3 plus.

So, both of them are independent in nature and during the formation of the code, we take the help of the protein and it is getting entered. So, at the same time another complex process how we get the corresponding iron from that particular code which is also very important; the way we analyze the overall minerals for iron analysis. So, ready use of that iron; that means the dissolution. So, not only the formation or the storage of the iron but dissolution of the mineral code is important and which is controlled again by the protein; that means you have the regular supply of iron inside the code and if you allow some reducing agent to entered there and if we are able to reduce this particular Fe 3 plus from this particular code. So, they will be left away or they will be released as Fe2 plus and those Fe2 plus are utilized for the synthesis of some of the important molecules like your iron sulfur proteins or the N proteins.

Thank you.