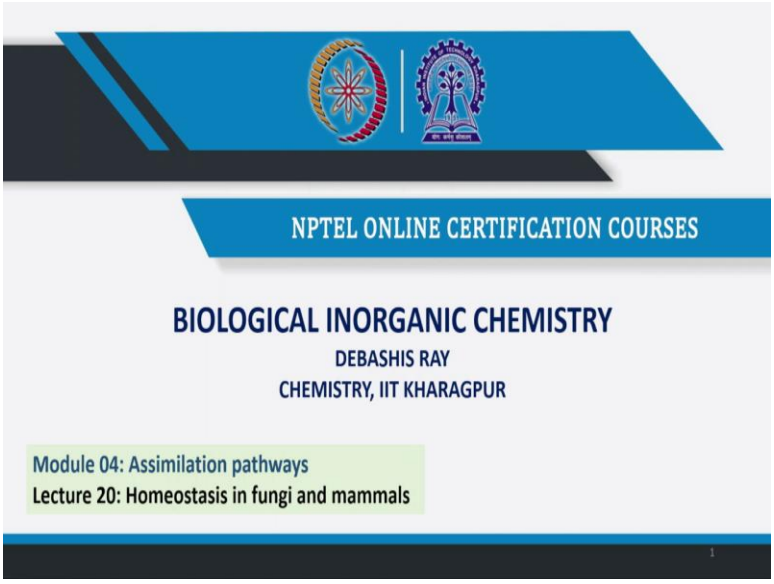
Biological Inorganic Chemistry
Professor Debashis Ray
Department of Chemistry
Indian Institute of Technology Kharagpur
Lecture 20
Homeostasis in fungi and mammals

(Refer Slide Time: 00:28)



Hello. Welcome back to module 4, where we are discussing about the assimilation pathways and in this last lecture of this module we will now move for the same homeostasis. Already we have defined it. We nicely know it now what is called homeostasis.

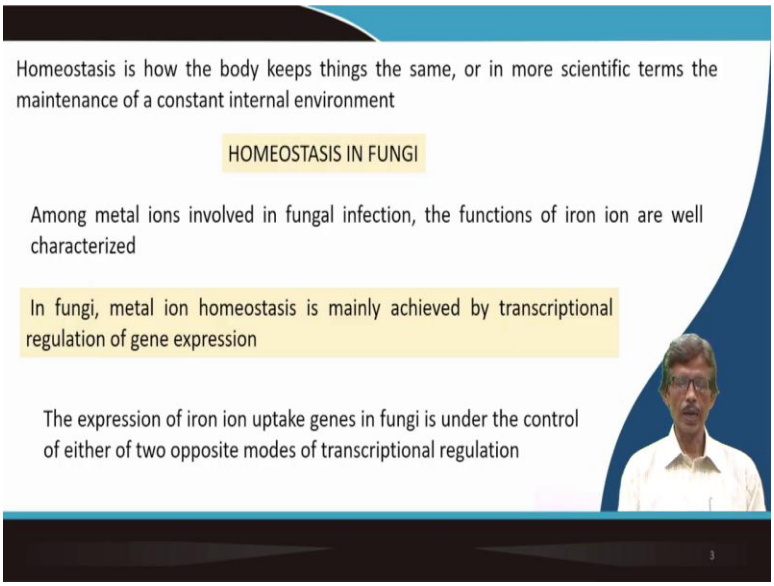
(Refer Slide Time: 00:50)

But now we apply for the fungus as well as the mammals and the human being, because these are very important to understand all these things in terms of the fungal metal ion homeostasis, how the different fungus can take up metal ion and whether they are dependent or not, we should also nicely know it. Then the transporters, not only that the metal ion transporters are available in the mammals or the human being, but these are also available for the different fungal environment, the bacterial environment and all these cases.

So, cellular homeostasis in human is also a very important thing and we will talk how we can assimilate from our diet, the iron. Then intracellular trafficking that means movement within the cell, intracellular trafficking within the cell, how iron can move from one point to the other. Then, connection to infection and diseases. So, these are very important thing.

The ultimate goal of ours is always that how metal ions are there and they are doing some crucial roles and playing some crucial roles and the x-ray metal ion as well as the deficit of metal ions, how it can go for the different disease conditions. So, our health, the human health is very much dependent on the proper assimilation or the proper homeostasis of these metal ions for our survival.

(Refer Slide Time: 02:09)



So, in is how we should know that homeostasis is, how the body keeps things the same. So, this is another way of defining homeostasis, because we want to retain the environment. It is not that you have taken your food in the morning and you are not taking food for the next four hours, but your proper assimilation of these in terms of food material as well as the iron environment or the iron concentration should be constant. So, body will always try to keep the iron concentration whether it is intercellular or intracellular, so intercellular or intracellular concentration that means, within the cell or outside the cell you always have some good iron concentration.

Also, if you are in a starving conditions that you are not taking food for 12 hours or 24 hours, what should be the condition, how the your typical biological system is getting disturbed and how the corresponding homeostasis of iron can play some important role to supply the iron from our storage that means from our liver the ferritins are there and from ferritins how we can supply those irons to our body.

So, in terms of more scientific terms, it is make keeping all the same is important, but maintenance a constant internal environment, whether you take your food or not you should be able to maintain a constant internal environment in terms of the metal ion concentration. So, in this particular class and all these classes, we are talking in terms of the homeostasis of the metal ions, not the other species. We are not talking in terms of homeostasis of vitamins, homeostasis of other proteins or homeostasis of the enzymes.

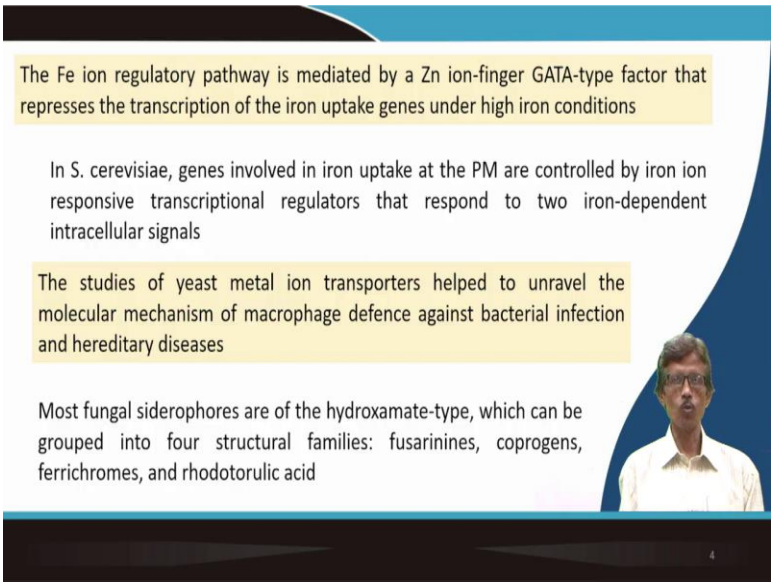
So, it is only, we are pinpointing our attention on homeostasis of metal ions like we are pointing the corresponding metal ions for their coordination chemistry. So, this is important. So, for the fungal thing, so we will typically see the fungal thing, because we all know particularly this is the corresponding in the month of July what we see that the season is rainy season to us and the fungal infections, we are facing fungal infections, the plants are also facing fungal infections and all these things, but everywhere the nice control or the minute control of iron is there.

So, for all these cases the metal ions involved in fungal infections the function of iron are well characterized, because we are studying for many years say the last 15 years or so the role of metal ions for all these infections and we are not still using because we consume the human being, as a human being we consume iron tablets, iron tonics when we are now we need more supply of iron, but we never put iron for the plants or the corresponding to stop the fungal infections you should go for the lower concentration of this iron such that the fungi are not getting that iron, the proper iron for their survival.

So, the metal ion homeostasis in fungi is mainly achieved again by transcriptional regulation of gene expression. So, these are the more complex biological terms, is my request is that do not worry about these complex terms. We are not biologists. We are not studying life sciences. We are studying only the biological part of your inorganic chemistry or the metal ion chemistry or the coordination chemistry. So, what you should think about, these are keywords and the definition what is gene expression and what is transcriptional regulation.

So, immediately if you see the statement that whether you require metal ion for these processes or not, so the transcriptional regulation of gene expression definitely needs the presence of the metal ion. So, for iron, the expression of iron, ion uptake genes in fungi is under the control of either two opposite modes of transcriptional regulation. So, if you can have the different types of transcriptional regulations, so you can have the two different modes and which are controlled by iron.

(Refer Slide Time: 06:16)



So, the pathway when iron is there and iron is mediating also, that particular pathway is being mediated by another metal ion dependent species which is your zinc ion-finger GATA-type factor. So, zinc ion-finger is a huge protein molecule. We know that zinc fingers, you can have the finger type of arrangement and the thiols are there which is immediately come and bind, trap the zinc centers.

So, that replaces the transcription of the iron uptake genes under high iron conditions. So, if you have higher concentration of iron, then something is happening in a different fashion and you should have the corresponding limit we should not have the higher concentration of iron, so the mechanism is going into the opposite direction. So, you will have the equilibrium condition at low iron concentration and at other case you have the high iron concentration. So, these two are matching through some mechanism again through some equilibrium process again.

So, S. cerevisiae genes involved in iron uptake in these at corresponding membrane, the plasma membrane, PM is your plasma membrane are controlled by iron ion unresponsive transcriptional regulators that respond to two iron dependent intracellular signal. So, when iron is present and iron is binding at some point and that sends the signal. So, if the genes are there and these transcription regulators are available, so they can sense or they can recognize those signals. So, recognition of those signals are important.

Until and unless you get the signal like when the train is crossing the signal, when the signal has a particular color only the train can cross. So, this is the same type of signal mechanism. So, signal is there. So, the presence of iron is getting detected and since the iron is present, so gene is there. So, gene will now respond in a fashion that such that it can finally synthesize the protein and the protein will finally grab that particular available metal ion.

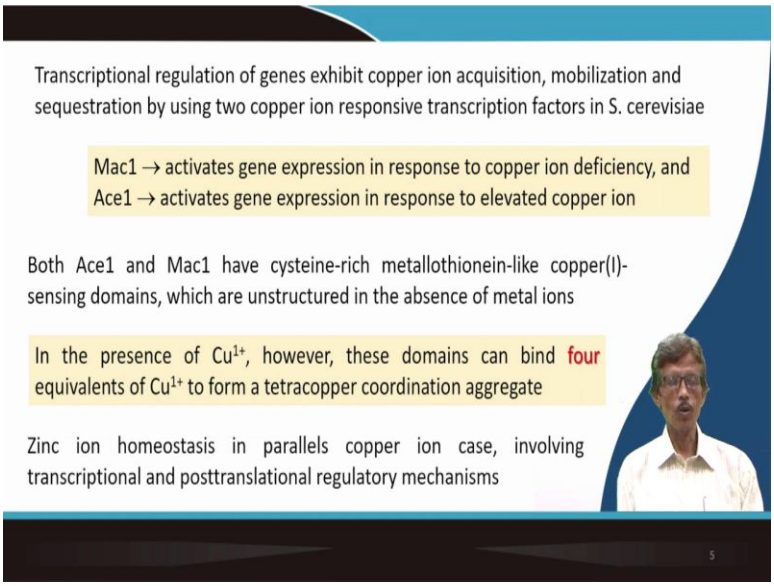
So, for yeast, we have seen earlier also in our many previous classes that yeast is also a very good model example of your fungus. The yeast metal ion transporters help to unravel the molecular mechanism of macrophage defense against bacterial infection and hereditary diseases.

So, these, why we are studying all these things such that we know a little bit about the role of iron in many, many bacterial infections, not only the bacterial infections, you can stop the bacterial, this particular infection by taking proper medication, but why it is happening? At other level how we can stop it? And sometimes the much more complex thing is the hereditary diseases, how we can stop this thing at this particular level also.

So, all these cases we know nicely that you have the hydroxamate function, the bidentate hydroxanate group is available for iron coordination, but can be grouped by four structural family, because the structures are different. The shapes of these molecules are different, big molecules are different. So, they are fusarinines, coprogens, ferrichromes and rhodotorulic acids. These are a little bit complicated names. Do not worry about the names. But these are, the biologists discovered all these things. They label these as and they put their name also.

But once you can understand that what is rhodotorulic acid, so this particular is the acid function. You can have some hydroxy functions and you can have some nitrogen donor group. So, in this big acid molecules you can have the donor groups, the atom donor groups or the ligand donor groups which can donate to the metal ion and it can coordinate. So, these we have already seen a little bit, but in future also we will see more of these groups, how they are coordinating to your metal ion centers.

(Refer Slide Time: 10:10)



So, this transcriptional regulation of genes also is responsible for your copper ion acquisition. So, we are not going far away. Most of these cases, we are talking some key words, this transcriptional regulation, transcription and all these things and the genes only. So, copper ion acquisition is there, already we have seen that, then the mobilization and sequestration, how, because you can have two copper ion responsive transcription factors in S. cerevisiae.

So, your model substrate is S. cerevisiae. So, S. cerevisiae is, we are studying on it, and then we can identify the transcription factors such that you can think or you can talk or you can consider the role of these transcription factors for copper acquisition for this particular biological species. Then, you level it. You see one Mac1 and Ace1. The Mac1 and what is Ace1. Then again it is controlling the transcriptional regulation and activates the gene expression.

So, genetically modified something we call the genetically modified seeds also for plants, the genetically modified brinjal and all these we know for so many years. So, these are the control of the corresponding gene expression until and unless we know the corresponding expression how gene is getting expressed, which will be responsible for your protein synthesis finally, so what are those things.

So, if you can able to identify, people have identified it, label it as Mac3, sorry, Mac1 and the condition is for copper ion deficiency. So, in case of copper ion deficiency we have to study Mac1. But when you have higher level of copper ions in the system, you have to study the Ace1.

So, you see now, so nicely we are now standardizing everything that you have the level of copper ions and we know that the corresponding homeostasis is controlling the iron concentration to a particular level which is the most standard level.

If you have extra iron it can be stored or if the mechanism is not there like iron that we can store iron in our body it should be removed from your system, from your body. But when deficiency is there, so the gene expression will tell you I have or the system has the corresponding smaller amount of iron. So, it will try to get more and more number of these metal ion for their survival. So, what are these? What are, these are nothing but the very simple molecule where you have the amino acid residue cysteine.

So, these cysteine amino acid residues in metallothionein like copper 1 sensing domains. The metallothioneins, we all know the metal ion is copper, thionin is the thiol we all know, the sulfur bearing molecules and the end part is the cysteine that means thiol Sh, after deprotination which will be S minus.

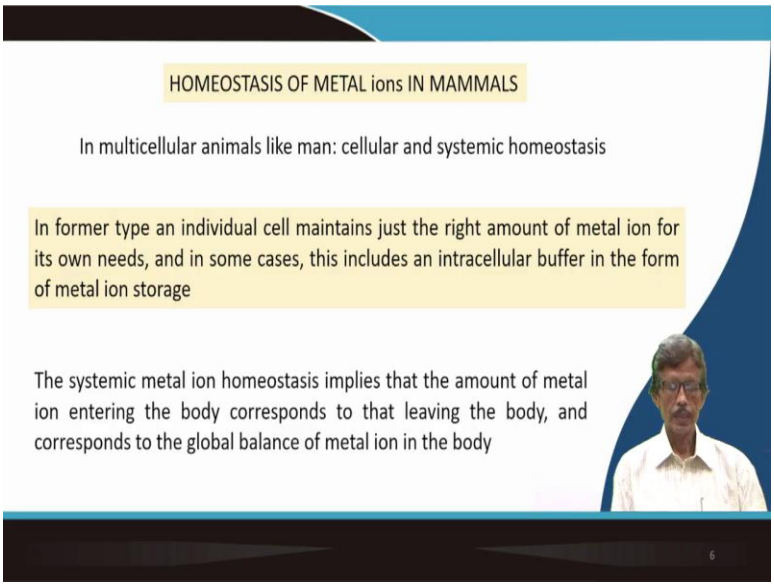
So, when they are coming and this particular reaction is very complex one if you try to do the reaction in laboratory for the complexation reaction of simple cysteine the amino acids that you can have in slightly basic condition in presence of say triethylamine you can react with some good copper source as copper 1 salt, you get it something which is the polymeric structure, because you have more number of these thiol groups will come and these thiol groups can not only coordinate to one metal ion center, but it can bridge by these two ends and bind two metal ion centers.

So, in a very uncontrolled fashion it can have, it can happen. But here systematically we can study this Mac1 and Ace1 for these copper sensing domains, which is unstructured in the absence of the metal ions. So, this is unstructured. So, it is very difficult to have this good structure. That is why we are talking about the x-ray structures, nice x-ray structure in presence of the metal ion.

So, metal ion not only helping the corresponding thing is in terms of your bio-coordination chemistry, but also the metal ions is also responsible for crystallizing your bigger biological molecules in good crystals. So, crystallization is getting help through the presence of these metal ions.

So, there are some domains and particularly these domains when copper 1 is present, these copper 1s can bind up to four equivalents of copper bond. So, you see immediately you get a tetrameric form. So, the motive is not a monomeric form, but you can have a tetrameric one, 1, 2, 3, 4, four copper centers are there and they are bridged by the thiols sulfur. And also for zinc ion homeostasis parallel to the copper ion case and which is also responsible for its transcriptional and post translational regulatory mechanisms. So, these mechanisms are also similar to that of your metal ions when your zinc is present.

(Refer Slide Time: 15:31)



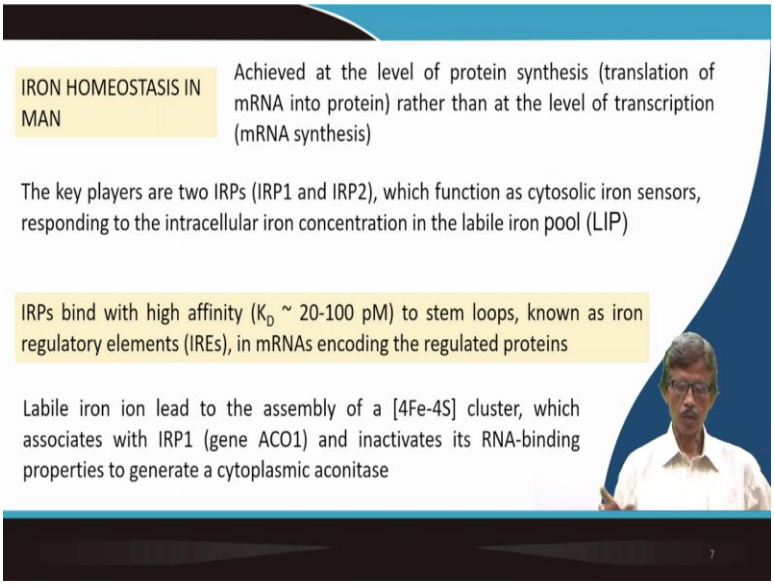
So, now, we will go for the corresponding one for the metal ion in mammals. How the homeostasis can take part in mammals, which we all know the much more complex multicellular species we are. So, the multicellular animals we are and we can have two types of homeostasis, therefore, one is, because we have studied a lot and we can now separate it out, one is the cellular homeostasis and another is the systematic or systemic homeostasis.

So, the systemic homeostasis is different from that of your cellular one. And the first case the former type an individual cell tried to maintain the right amount of metal ion concentration for its own needs, how much iron we need. So, that is followed to maintain a right amount of iron concentration. So, it will also include the intracellular buffer in the form of metal ion storage that if it is beyond that particular concentration you can have the metal ion concentration in solution which is functioning as a buffer.

We know that buffer is for your pH balance. When it is a buffered medium you are not able to change the corresponding pH drastically through the addition of base or to the removal of the base or the basic condition. Similarly, the buffered metal ion concentration is such that is difficult to disturb that particular concentration in that particular point. So, it also tells us that the amount of metal ion entering the body in our body corresponds to that leaving the body.

So, if the, from your food material the amount what we are consuming as iron, because your body is responsible for absorption and we will see also how it is getting absorbed in your intestine or in your gut that in that way you can have the total balance in our body. That means the right amount of iron you can have for all these cases in this.

(Refer Slide Time: 17:36)



So, what about iron? So, the this achieved and this particular one is achieved at the level of protein synthesis where you can have the translation of mRNA into protein. We all know the process is nothing but the mRNA translation for the protein synthesis rather than we see that during the mRNA synthesis, so during the translation process only not at the during the synthesis of mRNA and two of these species are IRP1 and IRP2 and they are functioning as the corresponding iron responsive proteins as your corresponding cytosolic iron sensors.

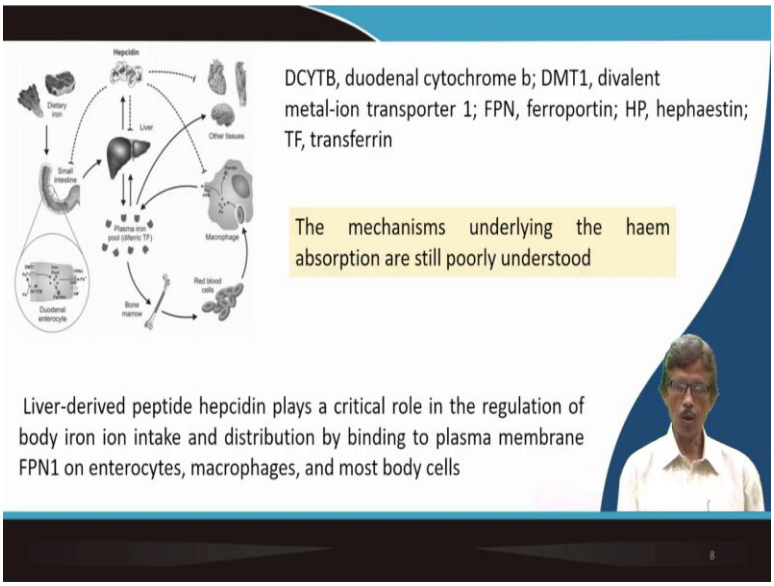
So, cytosol is there and cytosol should be able to sense the corresponding metal ion as iron and it can convey or it can that follow the corresponding signals. So, iron concentration in the labile iron pool. So, a large amount of iron you can have, but that particular concentration is must be

specified one. So, when they are there, this particular IRP is bind as a very high affinity in terms of its pM like your pH value. pH value tells us the hydrogen ion concentration. pM values are telling us the corresponding metal ion concentration in terms of iron if it is p Fe3 plus.

So, these iron regulatory elements are there in mRNAs and encoding the regulated protein that which particular protein will be synthesized in this particular condition. So, what sort of protein, already we know. We have studied in detail from our college days nowadays that the ferridoxin molecules. So, here we see now a detailed understanding of these things from the genetic level understanding will tell is this the labile iron pool the LIP can trigger the corresponding formation of your 4 iron ferridoxin molecules and which are associated with IRP1.

There are many IRPs we are just now talking IRP1, IRP2 and the gene is ACO1 and inactivates is RNA binding properties to generate the cytoplasmic aconitase. Aconitase are another variety of iron sulfur proteins. So, during this particular process, why at some point we are getting four iron species or four iron ferridoxin molecules, but there are some cases we can also find the corresponding three iron ferridoxin molecules or sometime aconitase where the citrate is binding. So, some other ligand part is coming and binding to that particular point.

(Refer Slide Time: 20:20)



So, now, if we see that we can have starting from your liver to that of the other parts the duodenal enterocytes, we are taking these as the food material. So, you have on the left top the dietary iron, the haem iron, the meat material and the vegetables, the carrots and all these, these are non-haem iron. So, you can have two different sources of iron and your small intestine is ultimately after digestion the peptidases and all these things are degrading the food material and it is reaching your intestine.

Then the duodenal enterocytes are looking for these irons. So, we will be talking in terms of corresponding the hephaestin, the hephaestin groups of molecules are there in your liver. So, what are the things which will be there starting from your red blood cells or red blood corpuscles even in the bone marrow, the bone marrow is producing your red blood cells. We all know that that we are synthesizing our, mostly our blood in our bone marrow.

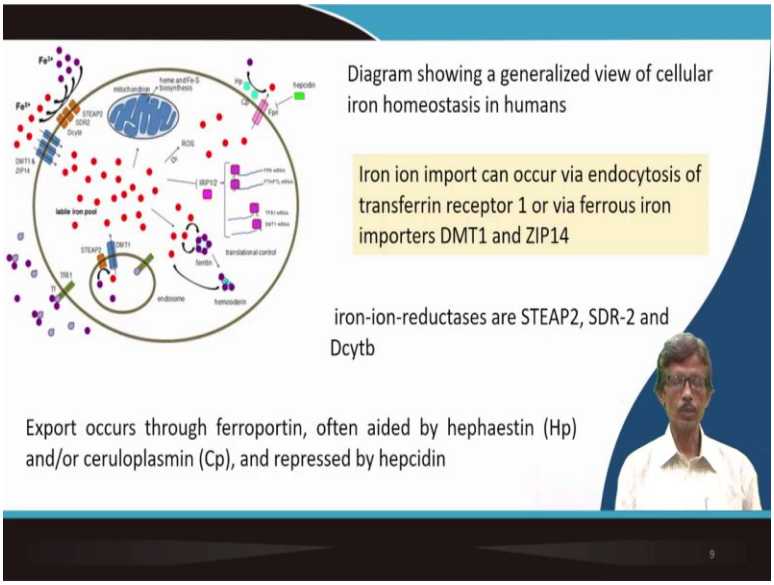
So, the duodenal cytochrome B is available for these all these functions. So, is DCYTB or cytochrome B, D cytochrome B, D is your duodenal, its origin is duodenal part of your small intestine. Then these are cytochrome b based that means again you have the iron. The presence of iron is there also. In cytochrome B you have iron.

Then divalent metal ion transporters, which can sense or which can think of that particular metal ion, then ferroportin already I told you that then hephaestin and the transferrin. Transferrins are the final thing. We will again discuss in detail when we talk about the individually the iron thing

in our body also, the hemoglobin, myoglobin, ferritin and transferrin. We will again come back there and talk about the transferrin, but not that this global picture in terms of your homeostasis, only we will look at the ferritin. But immediately you should be able to correlate it where we have learned the transferrin for the first time.

So, this mechanism of these all these things for the haem absorption and which is a difficult one rather than your free iron from the non-haem part is well known. So, it is poorly understood. So, the liver is producing these hepcidin and these liver derived peptide hepcidin plays a critical role in the regulation of body iron uptake and the plasma membrane FPN1 on enterocytes. Enterocytes are nothing but your corresponding duodenal enterocytes then the macrophages are there and most other body cells. So, all these different body cells will also be getting the take part in all these iron assimilation process also.

(Refer Slide Time: 23:11)



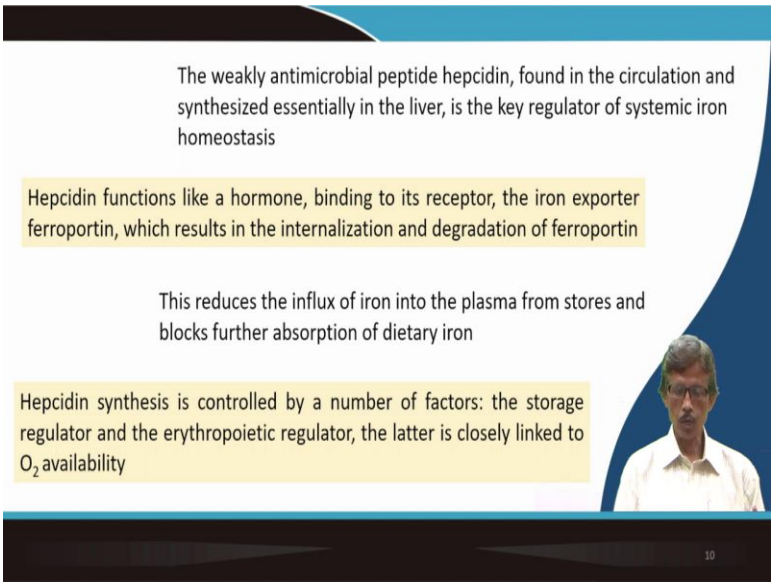
So, moving this iron from one point to the other, so it is iron is getting absorbed from food material to duodenum to intestine to the red blood cells and all these things, but how it is going inside the cell. So, if we talk two things, one is your cell and another is your whole body, you will now be confused. Do not get confused in this particular way. So, these are the very big thing. The whole body we are taking. And there we are talking about the iron homeostasis that how iron is getting distributed. Is the environment like your bigger environment is our body is

also environment, an environment of metal ions. So, how body will take care of all these things when we are taking the food material and all these?

So, similarly, within the cell also the cellular iron homeostasis is telling us that how in humans we can have these and the endocytosis we can have then the transferrin receptors 1. We will all talk again and again. But now we try to understand, we try to know all these DMT1, the divalent metal ion transporters,. So, is take care of the bivalent state of iron, so that is why it is the divalent metal ion transporters.

So, definitely, your question is very simple that if we can have iron 2 plus then only it can bind to your that DMT1. Then you have the reductases that means, if the iron 3 is there we have to reduce it to the corresponding one such that it will be sensible to your DMT1. Then the export, the export occurs through your ferroportin often added by hephaestin and the ceruloplasmin. All we know the ceruloplasmin is a copper bound metallobiomolecules which is also there and which is also help us in understanding something in terms of this electron transfer or other thing.

(Refer Slide Time: 25:05)



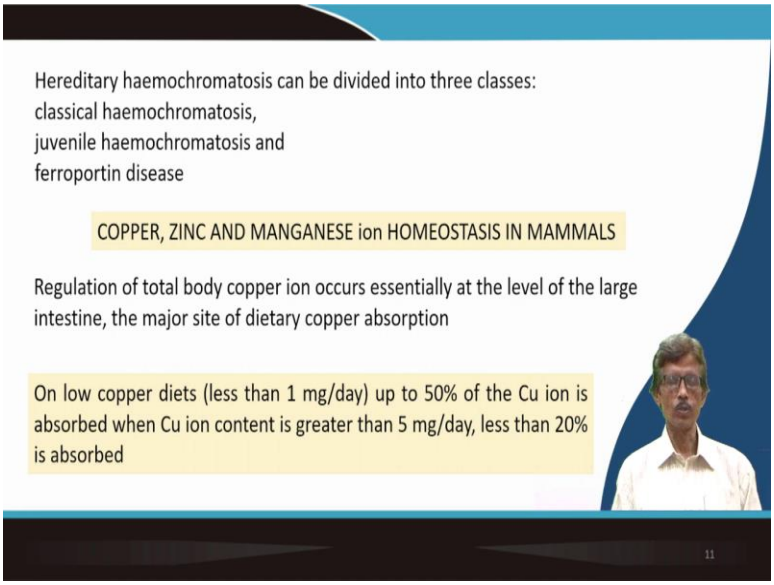
So, this weakly antimicrobial peptide hepcidin found in the circulation and they are synthesized using the liver and the systematic iron homeostasis is taken care up by the liver itself. And it functions like how it functions the peptide molecule hepcidin. You can go to your simple Wikipedia page type hepcidin only that hepcidin will give you some brief introduction for that what is that hepcidin. So, you just have a note also for that what are these hepcidin function and

these thing, because we are learning the name, but the actual function we are not studying all these things, but we are talking in terms of its dependence and relationship with the metal ion.

So, degradation of the ferroportin also the iron export protein is also there. And this reduces the influx of the iron into the plasma from the stores. If you have the iron stored in the ferritin molecule and when the influx is taking place and the signal is also going in this particular fashion that it will telling the corresponding small intestine that do not go for absorption of more iron. So, iron will not be absorbed more. So, that particular signaling business is very interesting that that signaling only will control that whether you will have more iron absorption in your small intestine. So, you can control.

So, all these signaling mechanism and all these it is not, that is not voluntary, that whatever food material you will be taking all will be responsible for your iron absorption. Then this hepcidin synthesis is controlled by a number of factor, one is your storage regulator and the erythropoietic regulator and which one is lastly related to your oxygen availability that means O2 available which is nowadays is also very much important and is a thing of concern also because this O2 availability is due to the corresponding redox cycle or the redox reactions.

(Refer Slide Time: 27:09)

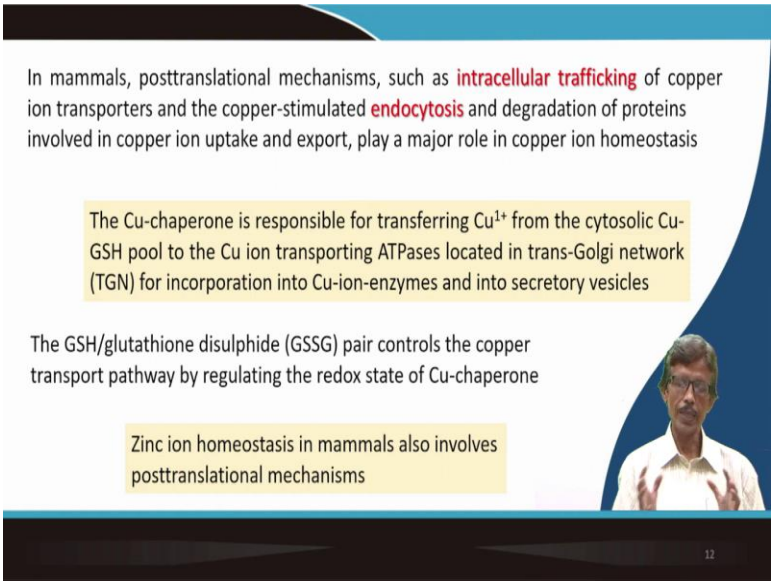


So, one such disease related to your corresponding haemochromatosis is one is classical type, another is juvenile type, another is corresponding related to the ferroportin disease. So, once you know there is improper assimilation or improper homeostasis then we can correlate that we can

have some diseased conditions. So, like that iron you can have the other three metal ions we can talk about or we can think about in a similar fashion. And now at the level of the body copper occurs essentially in the large intestine not at the small intestine. So, the location is different.

So, we can compare it now, and the diet what you are taking is 1 milligram per day. When you are taking a less amount of copper in your diet, your body mechanism is such that it will try to gather more and more than 50 percent of the absorption can take place. So, 0.5 milligrams of that can be absorbed at a time. But when you are taking higher copper rich sample or the food material, your absorption will be less. It is 20 percent. So, 20 percent of this is only 1 milligram. So, up to 0.5 milligram to 1 milligram it will be the range for your copper absorption in all these cases.

(Refer Slide Time: 28:18)

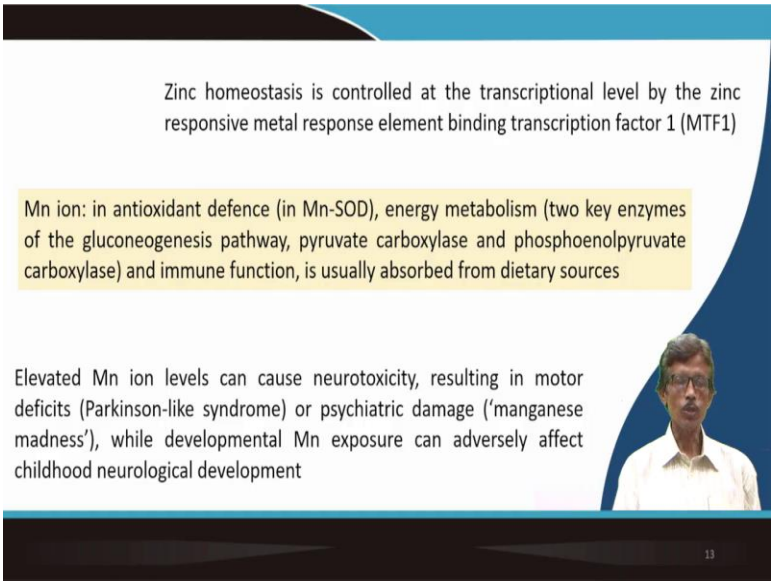


So, in mammals, we can have the intracellular trafficking and we can have the endocytosis, but that is copper stimulated and degradation of the proteins involved in copper ion uptake and export. So, how much copper you are taking up and the export mechanism is responsible for your endocytosis behavior and which is also giving contribution to your copper iron homeostasis. Then one such molecule is copper chaperone or chaperone what as you say is responsible for transferring copper 1 plus in cytosolic copper glutathione pool.

We all know the glutathione as well as glutathione disulphide which are having the sulfur like cysteine and cystine. Since copper has higher affinity for sulfur coordination so all these things,

so you have to identify not only copper ion in the solution or the cytosolic solution, but also you have to find out the corresponding GSH or GSSG. Then like copper you can have also the zinc ion homeostasis and also involve in that particular genre or domain or particular time where you can have the post translational mechanisms which is operating.

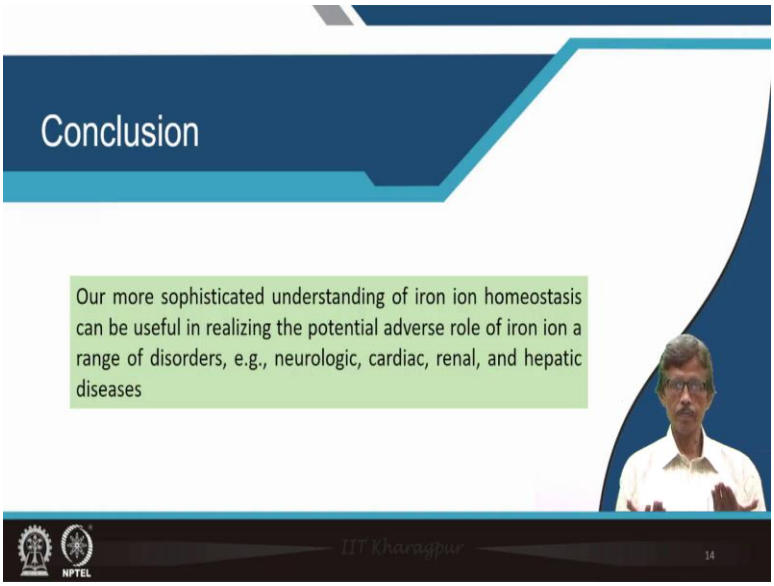
(Refer Slide Time: 29:34)



So, this zinc homeostasis is controlled by translational level and there at some transcription factors. Then finally we will see how manganese ion manganese ion, manganese ion is an antioxidant defense mechanism because we all know manganese SOD, which is very important nowadays. People are talking about genetically modified plants also. People are planning for that. But before that we should also know about how much manganese can play some role for our dietary source whether we should take some food material which is rich in or deficit in manganese.

So, elevated manganese, this also deadly. That is why we can have the typical balance. Why we should be very much careful in knowing all these things is not that that you are thinking that zinc is bringing your immunity and all these things. So, you take up continuously zinc is very bad. Similarly, for the elevated level of manganese we do not know your body mechanism is such that you can have some elevated level of manganese, but this can give you some diseased conditions, the Parkinson like syndrome, manganese madness and all these things can happen and different, due to different neurological development from your childhood time also.

(Refer Slide Time: 30:42)



So, in conclusion, what we can see now or say now is that our sophisticated understanding which starts from your iron ion homeostasis can be very likely, is very easily realizing the potential adverse role of iron ion in range of disorders, in case of your diseased conditions. Why we should study these, all these role of iron and zinc in translational factors and all these big, big thing, very complicated? People can think of particularly people having background in coordination chemistry and who are studying the chemistry.

But it is your duty that you can only think of the metal ion. No other people can think of much more about the metal ion and its coordination chemistry as well as the chemistry related to the metal ion. So, we see the range of disorders can be only be monitored or only can be considered if the you can find out the disbalance or imbalance, what you can think of, okay, due to iron imbalance, you can have the neurologic disturbance, you can have the cardiac disease, you can have the renal as well as the hepatic diseases. So, all these things we can systematically see and learn in terms of the metal ions.

(Refer Slide Time: 31:54)

So, human iron metabolism is also a very good part in Wikipedia and you can go through that and also the book every time I am giving you the reference. So, thank you all for your presence.