Overview and Integration of Cellular Metabolism

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Week 10

Lecture 46: Heme Metabolism – I (Heme Synthesis and Regulation)

Hello everyone, welcome back to your classes on Overview and Integration of Cellular Metabolism. From today's class we will be starting heme metabolism, specifically today we will be discussing about heme synthesis and regulation. So, you might wonder why we are studying heme, this should raise your question whenever you see a topic you must ask why. Why we will be studying the structure of porphyrin and heme in this class, why do we need to structure I mean need to know the pathway of biosynthesis of heme, why do we at all need to know the regulation of heme synthesis. This is because most of you already know that this has got a big role to play in red blood cells ok. So, when we are discussing RBCs you should know first a basic thing about RBCs is a very basic knowledge that RBCs are about a biconcrave non nucleated structure which have got 7 micron right structure and their half life is 120 days this you already know from discussion regarding HbA1c that RBC of lifespan is 120 days and that is why it has got some importance right.

So, this mature RBC do not have nuclei and hence they do not undergo the pathways that are there inside organelles like mitochondria and nucleus. However, RBCs do have cytoplasm and do they do have the cytoplasmic enzymes. So, they mainly depend on glycolysis for energy and production of 2, 3 BPG this is all discussed these are just revision lessons about RBC right. You can see unknowingly you know so much already about RBC and you also know that the NADPH that is required in RBC to fight off those culprits that were trying to degrade the RBC membrane they come from the HMP shunt pathway right.

So, what is the main role of RBC why do they look red they mainly carry hemoglobin and for that heme synthesis is one of the main role of red blood cells right they form you already know they form they are forming the bone marrow and this reaction needs many compounds or many elements many nutritional support they are some amino acids they are iron, copper, vitamin in the form of folic acid, vitamin C, vitamin B12 and vitamin B6 and pantothenic acid. So, all of these are needed for formation of blood and they are

known as hematinics. So, whenever we utter that are hematinic it means any drugs that are you prescribed to form blood ok and multi I mean these are specifically prescribed during pregnancy. So, blood forming syrups all these things contain not only iron, but multiple all of these are mixed ok. So, with this basic knowledge of RBC we should also know some basic knowledge about the structure we should also have some basic knowledge about the structure of hemoglobin.

We already know it is an iron containing conjugated protein that has got heme and globin. So, heme is the actual prosthetic group and the protein is the globulin. If you look at the structure it is a tetrameric protein 4 subunits are present in each subunit having a prosthetic heme group and the globin polypeptide. 2 alpha and 2 beta forms the 4 tetramers ok. The molecular weight is 67 Kd kilo Dalton or 67000 Dalton right and each gram of hemoglobin mind it hemoglobin is in gram per deciliter they contain 3.

4 mg of iron. So, what why do we need to know so much about heme because heme is not only present in hemoglobin it is also present in myoglobin cytochrome peroxidase enzymes like peroxidase catalase tryptophan pyrolase and nitric oxide synthase ok. So, very very very important these enzymes also contain heme and you may get an multiple choice question on MCQ where each of these may be given as an answer which you can choose as a direct answer as an all as an all except type of question. So, first things first heme is produced by the combination of iron with a porphyrin ring it is similar in which the counterpart that is chlorophyll in plant is also a combination of a magnesium ion and a porphyrin complex right. And you know the scientist who actually received Nobel prize for his work on hemine and chlorophyll is actually Hans Fisher he was a German scientist who discovered the role of both chlorophyll and hemine.

You know he was a so much dedicated scientist that he dedicated his whole life into finding all of this and his whole life entire world in Germany was actually destroyed during the end days of world war 2 and he committed suicide. So, very sad story, but it is a very inspiring that he already got Nobel prize very very very vital in this context of our discussion. Regarding the structure of heme it is a derivative of porphyrin. Porphyrins are cyclical compounds formed by for fusion of 4 pyrrole ring. What is pyrrole ring? This is how a pyrrole ring looks like and when we abbreviated by omitting the carbon and hydrogen it is often signalled as this right.

And these 4 pyrrole rings are linked by methemyl bridges and since iron atom is present it is also a type of ferro protoporphyrin ok heme is a ferro protoporphyrin will be discussing what we mean by protoporphyrin ok. So, then regarding the nomenclature of pyrrole rings they are named as alpha beta gamma and delta ok based on the sequence. So, this is alpha beta gamma and delta and generally we often I mean for simplicity of

understanding we often represent this as in this structure ok where the sides for attachment of side chains are represented by the corner of this huge plus sign ok. So, you should get used to this type of nomenclature, but in reality you should have in mind that in reality that is 4 pyrrole structure which are connected by methemyl bridges. So, they the porphyrin ring this is the porphyrin ring ok this porphyrin ring are of multiple types multiple subclasses the first one that is one ok they have gotten arrangements symmetrical arrangement of substituent groups we will be seeing what are the substituent groups very soon.

So, number if you just number it like this way right. So, if you just notice the number of groups the groups that are present in 1 3 5 and 7 and 2 4 6 8 if they are symmetrical then it belongs to first series. If they are asymmetrical then it is known as series 3 right and type 3 is the most predominant variety in biological system. There are also multiple types that we do not need to know that if you just know type 1 and most importantly 3 you are good to go. It is also called series 9 right because Fisher the discoverer the pioneer in this porphyrin chemistry has placed as the 9 series in 15 possible isomers ok.

So, if we look at the all the possibility the original discoverer placed this our product of importance in 9 ok. So, it is also it is also referred as 9 in some text books ok to respect the discoverer, but you should always know type 3 is the most predominant and this is the most interest for us ok. So, what are the substituent groups what groups can be placed in 1 to 8 ok these are the ones propionyl acetyl methyl and vinyl they are often abbreviated as P A V and M we will see very soon right. So, depending on the order of substitution from 1 to 8 this is the nomenclature and there can be again multiple isomers or multiple forms of this porphyrin to us for heme synthesis these are the intermediates that are required and these are the substitutions. Again it is almost an impossible task to remember, but you still need to remember few which I will tell you very soon.

So, this is the structure of heme in which you should remember the substitution in order to get some good example this is a nice to know area it is not a must know area, but you should know right. So, the heme in heme the substituent groups are methyl vinyl, methyl vinyl, methyl propionyl, propionyl methyl you can remember it in any way you can remember M V M V M P P M it is up to you, but you should remember the final structure of heme and it is substituent groups right. So, heme is actually can be synthesized in almost all tissues in the body right in case of RBC it is synthesized not the mature ones, but the immature in bone marrow that is in the normoblast is actually the intermediate normoblast where hemoglobin first appear you already might be knowing that from your physiological knowledge right. And if you look at the pathway the pathway is partly happening in the cytoplasm and partly in the mitochondrion this is the reason why when RBCs are fully matured they do not have mitochondrion heme

synthesis cannot takes play cannot take place because there are few mitochondrial enzymes that are also essential in heme synthesis. So, this is the step 1 we have already discussed during glycine metabolism how heme heme was being synthesized as an intermediate.

So, succinyl CoA and glycine under the action of the enzyme deltamino levalinate synthase it is forming deltamino levalinate there is an keto adipate intermediate, but ultimately this is the final reaction right. So, succinyl CoA glycine forming ALA this is also abbreviated as ALA or ALA one very important thing to notice this requires pyridoxal phosphate as coenzyme and this is the reason why in vitamin B6 or pyridoxal deficiency there will be defectin heme synthesis and there will be anemia ok. This is the rate limiting enzyme if we control the activity of this enzyme heme synthesis will be affected and regulated and controlled right. This is the major pathway that is happening in higher animals, but in bacteria and plants an alternate source of ALA is also glutamate right it can be directly synthesized from glutamate via a semi aldehyde form ok. So, in the second step what is happening two molecules of delta amino levalinic acid are joining together right and it is acted upon by a dehydratase enzyme.

This enzyme has got multiple names we can call it ALA dehydratase depending on the acts action on the substrate or we can also name that enzyme depending on the product is forming that is porphabilogen synthase it is the same thing and leads to the formation of porphabilogen ok. So, this is the second step in which two molecules why it is showing 8 and 4 because in reality we need four molecules of porphabilogen. So, actually at one reaction level two molecules of ALA are combining one molecule of water is lost and porphabilogen is formed, but in reality eight molecules of ALA are forming four molecules of porphabilogen this is the step 3. So, what happens after formation of porphabilinogen? So, four molecules of porphabilinogen results in the formation of the first porphyrin of the pathway that is europorphyrinogen ok. Europorphyrinogen is a cyclical compound this is not yet cyclical ok this is you can see the enzyme europorphyrinogen synthase actually the four porphabilinogen combines in such a so that a linear tetrapyrrole ring is formed it is not yet cyclized ok this is known as hydroxymethyl bilane or pre europorphyrinogen HMB this enzyme is also often known as hydroxymethyl bilane synthase HMB synthase ok.

We will be discussing in detail again in porphyrin so do not worry. So, europorphyrinogen synthase it forms a linear compound which undergoes spontaneous cyclization to form europorphyrinogen 1 ultimately that one is a symmetrical porphyrin ok what happens by europorphyrinogen cosynthase it is converted to europorphyrinogen 3. So, everything is happening in one step. So, this linear compound pre europorphyrinogen is being converted to europorphyrinogen 1 spontaneously and then

by the action of europorphyrinogen 3 cosynthase it is con I mean formed converted to europorphyrinogen 3. So, europorphyrinogen 1 is basically the symmetric isomer ok in which everything AP AP AP AP is present in 1 to 8 in symmetrical manner and only thing that is different in europorphyrinogen 1 and 3 is the interchange of this A and P if it goes here it becomes third isomer.

So, all this is all what is happening in this reaction. So, ultimately we are getting europorphyrinogen type 3 ok. So, what happens when this fusion occurs there might be many 1 isomer there may be multiple 3 isomers ok because not all the 1 isomers are converted to 3, but from now only series 3 are further utilized ok. The pyrrole rings will be joined by methylene methylene bridges which is derived from the glycine ok. And we should know that porphyrinogen are actually colorless still we are in porphyrinogen these compounds are in porphyrinogen, but once they are oxidized we will be reading very soon they are known as porphyrins which are colored compounds ok.

So, in the next step what happens europorphyrinogen loses carbon dioxide to form coproporphyrinogen europorphyrinogen 3 forms coproporphyrinogen. What actually happens the acetate groups are decarboxylated to form methyl group there are 4 acetate groups CH 3 COOH basically CH 2 COOH the carbon dioxide if it goes it will form it will CH 3 will remain and that is exactly what is happening it is very easy to understand that molecular ok. You should know molecular oxygen is required in this reaction. So, oxygen might come anyway so you can see hm. In the next reaction what happens further metabolism takes place in the mitochondria.

So, from here we will go into the mitochondria right coproporphyrinogen is oxidized to protoporphyrinogen 3 by action of coproporphyrinogen 3. So, oxidase ok this enzyme specifically acts on type 3 series and not on type 1 and 2 propionic acid side chains are oxidatively decarboxylated to vinyl groups ok and this reaction also requires molecular oxygen. So, what happens oxygen is coming and CO 2 is produced and protoporphyrinogen is formed from coproporphyrinogen by action of coproporphyrinogen oxidase ok. Well in this reaction oxygen is actually not required only CO 2 groups are removed here oxygen is required we need oxygen in order to convert the propionic acid to vinyl group whereas, over here we can just remove carbon dioxide to form methyl groups alright.

So, be very careful. The next step is actually generation of protoporphyrin. So, from protoporphyrinogen it is getting converted to protoporphyrin and this reaction also requires molecular oxygen ok it is also happening in the mitochondria and as per the discoverer he named it protoporphyrin 9 and it is in this reaction that the methylene bridges are oxidized to methenyl bridges ok this is a double bond incorporation over here

ok. So, we are getting protoporphyrin we have almost reached the last step because in the last step this 4 I told you that heme is actually ferroproporphyrin. So, now iron it needs to get inside. So, iron by the action of the enzyme ferrochilates or hentzyn phase is getting accommodated and protoporphyrin forms heme alright.

So, if we look into the valency of iron it is actually attached to the 4 nitrogens ok and the first atom nitrogen atom histidine ok 5 nitrogen atoms and the remaining valency is satisfied with either water or oxygen atom depending on the oxygen carrying I mean since the role is carrying capacity I mean the role of iron is to carry oxygen in heme. So, that valency the last valency satisfied by oxygen or water right. So, this is actually the entire pathway of heme synthesis where all the intermediates have been shown this is the number. So, we started with 8 we ultimately needed 4 because 4 condenses into 1 which ultimately undergoes various changes to form a protoporphyrin which by the action of the ferrochilates enzyme incorporates an iron to become heme. Again the name of the intermediates are necessary the enzymes are necessary, but the structures are not only the final structure of heme is important where we need to know the substituent groups ok.

And if we look into this pathway linearly this is how it happens it starts from succinyl cloyetoglycine this is the rate limiting enzyme and thereafter all the reactions are taking place and by the action of final ferrochilates enzyme we are getting heme. We will again be revisiting this pathway when we are discussing degrade I mean disorders of heme synthesis that are known as porphyrias ok. So, if we look at the compartmentalization it is nothing, but the reactions that are happening partly inside mitochondria. So, first second third fourth fifth sixth seventh you can see 2 3 4 is happening in cytosol and 1 5 6 7 is happening inside mitochondria right. So, be very careful about which enzymes are present where.

We should know that when this iron is generated in ferrous form if it is oxidized to ferric form it loses its capacity to carry oxygen it is known as hematin. Here heme is red in color, but hematin is dark brown why do we need to know about hematin number 1 Hans Fisher discovered it number 2 hematin plays a role in regulation of heme synthesis. So, we finally, shall discuss regulation of heme synthesis there are multiple mechanisms the first mechanism is the rate limiting enzyme deltamino leveronic acid or alla can be feedbackly inhibited by heme itself ok repression mechanism and he is a co repressor not only that it can be allosterically inhibited by hematin ok. Next the compartmentalization of enzyme also helps in the regulation I told you the rate limiting enzyme is present in the mitochondria and 1 5 6 7 taken place in the mitochondria and 2 3 4 are taking place inside the cytoplasm. So, because all of the enzymes are not present together depending on the required amount of heme synthesis thus enzymes can be increased or decreased in the various fractions of the cell and the synthesis can be regulated ok.

Now the action of drugs drugs like barbiturate they induce heme synthesis they increase the rate of heme synthesis because barbiturate required cytochrome. I told you in the first slide heme is not only present in hemoglobin it is present in myoglobin cytochrome etcetera and 1 very important product that is cytochrome p 450 enzyme system is required for metabolism of barbiturate. So, out of the total heme produced two third is actually used for cytochrome p 450 production only one third is used for the production of heme right and how it can be inhibited it is inhibited by lead specifically the enzyme ferrocilatase and allerdehydratase they are inhibited by lead ok. So, lead poisoning is to anemia and heme synthesis ok. Next is INH it is an anti tubercular drug ok anti tubercular drug INH it also decreases the availability of heme and may actually actually it inhibits vitamin B 6 availability pyrodoxal phosphate.

I told you pyrodoxal phosphate is a very important cofactor of alla synthase alla enzyme deltamyl levalinic acid synthetase. So, by regulating enzyme affection it also affects heme synthesis. Higher cellular concentration of glucose prevents induction of alla synthase this is the base. So, this is the basis of administration of glucose to relieve the acute attack of porphyria we will be discussing in detail in the next class where we will be discussing the porphyria right and we should also know that alla synthase has got both non erythroid forms in it is I told it is present in RBC and also in hepatocytes right. So, erythroid form is actually alla synthase 2 and it is not induced by drugs that affect alla synthase 1 right and this erythroid form is also not subjected to be feedbackly inhibited by heme.

So, very important we can also always carry on production of blood I mean heme in our bone marrow ok it is not feedbackly inhibited. So, these are the various regulation mechanism of heme. So, to conclude we have discussed in detail the structure of heme the pyrrole ring the porphyrin ring we have discussed what are the intermediates of heme synthesis, but I already told you the structurally this will not be important, but the final heme molecule is very important we have discussed the steps of heme synthesis we have discussed how the heme synthesis is regulated. So, these are my references for this class and I thank you for your kind attention.