Overview and Integration of Cellular Metabolism

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Lecture 11: Hexose Monophosphate Shunt : Regulation and Significance

Hello everyone, welcome back to your course on Overview and Integration of Cellular Metabolism. We are continuing with the carbohydrate metabolism in the last lecture, we learnt about the various phases of hexos monophosphate shunt and in this class we will be covering these following concepts ok. So, these two are covered in the last class the hexos monophosphate shunt pathway and how it can be divided into how it can be remembered easily by splitting the pathway into two phases that is oxidative and non oxidative phase ok. So, today we will be continuing with the regulation of HMP shunt, how the whole thing is regulated, the rate is controlled of the HMP shunt pathway and also the metabolic and clinical significance of HMP shunt pathway, why is it so important for us ok. So, this you already remember ok from the last class this is known as either pentose phosphate pathway or Warburg pathway or phospho gluconate pathway and the importance was it is an alternative pathway to glycolysis and TCA cycle for oxidation of glucose right. And I also told you the HMP shunt is actually more anabolic in nature, why this is important because you already discussed in the regulation of enzyme class knowing whether a pathway is anabolic or catabolic will help us to predict how it is regulated.

Mind it even though it is a pathway of oxidation of glucose means ultimately the glucose molecule is broken down. However, the line is very important read by the lines it is more anabolic, there is a catabolic component, but it is more anabolic in nature because of the production of component like pentose and HNADPH which are further used to synthesize more components. So, this is the pathway which has been discussed in detail in the last class. So, if you cannot recall it right now it is a very good time to pause this video, just recap the earlier class and then come back here again ok.

So, the question arises in this class is how it is regulated ok means how the rate of HMP shunt is controlled very important. The one and only answer is by the cellular concentration of NADPH the reaction of the enzyme glucose 6 phosphate dehydrogenase is controlled. See this is the cycle glucose 6 phosphate dehydrogenase was the first step

G6PD the action of this enzyme is controlled. So, if we can control the action of this enzyme the entire HMP shunt pathway the flow will be controlled and who can control this G6PD that is NADPH or even NADP plus it is the NADPH to NADP plus ratio. So, you can get any option NADP is important mind it is not NADH, NADH is very different from NADPH we are looking for the P over here when we are dealing with HMP shunt.

So, whether it is an important product or whether it is regulated regulation NADPH is your answer of choice ok. And what is the rate limiting step or the most important step which with which we can control HMP shunt that is glucose 6 phosphate dehydrogenase either GPD or G6PD this is the answer all right. So, since it is an anabolic process now we extrapolate our concept that we learned in the very second lecture that is regulation of enzyme action by covalent and hormonal modification. So, when a process is anabolic what hormone in what form will help its progress anabolic insulin dephosphorylation the rule applies here also. So, the synthase of glucose 6 phosphate dehydrogenase that is from glucose is actually induced by the increased insulin is to clog on ratio means more the insulin is the more will be the pathway.

When is the insulin level high after a carbohydrate rich meal. So, when body is rich in carbohydrate insulin level goes up insulin helps in this pathway ok. So, insulin will induce glucose 6 phosphate dehydrogenase and overall increase the rate of the pathway ok. So, this non ox and this was about the oxidative phase because in oxidative phase glucose 6 phosphate dehydrogenase acts right and what about the non oxidative phase. So, in oxidative phase who was getting produced mainly carbon dioxide right and in the non oxidative phase various pentoses were produced.

The non oxidative phase is regulated by the requirement of pentoses how much pentose is required for the body why pentose is required for the body it is required for production of DNA and RNA. If we need pentose we need more source of pentose that is HMP shunt and then also HMP shunt will be up regulated. When there is no requirement of pentoses when there is less insulin high glucose on the process will be inhibited it is as simple as that. Now, we already have discussed briefly the significance of HMP shunt that is it plays a major role to produce two most important compound number one pentoses or pento sugar for formation of DNA and RNA as well as NAD pH ok. I already I also told you something in last class it might appear trivial it is very, but it is very important that is 95 percent or more than 95 percent of the body's NAD pH is produced by hexos monophosphate shunt and less than 5 percent is produced by malic enzyme, malic enzyme not malate ok malic enzyme it is quite different.

So, what is the importance of pentose? The hexose are converted to pentose we already

know how hexoses are rearranged to pentoses and it is important in synthesis of DNA and RNA deoxyribonucleic acid and ribonucleic acid the very fundamentals of like the very fundamentals by which DNA and gene and chromosome are made of right DNA makes chromosomes after supercoiling. So, those ribonucleic acid are derived from ribose which is a pentose produced from HMP shunt. Next what is the importance of NAD pH? NAD pH has got a lot of functions in the body right way we can see we will first I mean enlist what are the function then we will delve into small details which is required for you. So, number one it is required for biosynthesis of fatty acid and steroids we will be discussing them later during the course when we are dealing with lipid metabolism. Apart from that NAD pH is used in synthesis of certain amino acid involving the enzyme GDH or glutamate dehydrogenase we will cover in detail when we are dealing with protein and amino acid metabolism, but for now you should note down this example.

So, that you can recall later because everything is linked right we will also tie up everything during integration of metabolism session anyway. Next most important activity of NAD pH is for a defense mechanism I told you during the course outline we need to learn metabolism to so that I mean cell why metabolism is important because cell needs metabolism to defend itself against external stimulation external infection this is the key area. Because NAD pH or nicotinamide adenine dinucleotide phosphate the reduced form NAD pH is actually responsible for production of reactive oxygen species by macrophages which help in killing the bacteria. How does it happen you see the phenomena is known as respiratory burst ok. So, respiratory burst is the phenomena by which multiple compounds starting with NAD pH are involved in production of reactive species or ROS which ultimately acts as a defense mechanism to kill the bacteria.

See how this occurs we can get NAD pH from glucose this is the HMP shunt pathway ok by the enzyme glucose 6 phosphate dehydrogenase on NAD pH the enzyme NAD pH oxidase acts. So, when NAD pH oxidase acts it is reduced to NAD P plus this hydrogen this is the nascent hydrogen is used I mean this electron ok reduction oxidation is an exchange of electron this actually is used to convert oxygen to superoxide radical O2 minus dot this is superoxide radical ok. So, NAD pH oxidase enzyme is actually producing superoxide radical which is further acted upon by the enzyme SOD or superoxide dismutase short form is SOD. So, SOD acts on O2 minus that is superoxide radical to produce H2O2 or hydrogen peroxide right. Next in next step what happens myeloperoxidase or MPO acts on this hydrogen peroxide in presence of fluoride ion to produce hypochlorous acid.

Hypochlorous acid or H it is also denoted by HOCl, HCl or HOCl this hypochlorous acid as well as the superoxide radical and even in some cases the H2O2 can directly

attack and damage the bacteria or the foreign invaders ok. So, it helps in production of reactive oxygen species and it is a reactive oxygen species which are arrows by which a cell shoots the arrows at those foreign bacteria and they die as simple as that. But you know what happens there is a I mean problem to the whole thing because once the bacterias are gone ok arrows are short 100 arrows have been shot 50 have killed the bacteria. So, what will happen with the rest 50 if left just like that if ROS or reactive oxygen species is just left roaming in the system what will it do it will attack our own RBC membrane. So, these free radicals that are generated needs to be scavenged needs to be taken care of who does this.

First of all why it is important to scavenge the free radicals because as I told you the free radicals are constantly produced inside the cell and if left unchecked they will destroy the DNA protein fatty acid and all biomolecules will be destroyed. It will create a chain of cellular oxidation reaction it will lead to lipid peroxidation the entire cell membrane will be damaged cell membrane DNA RNA everything. So, this free radical is actually there is a mechanism by which this free radical can be checked the progress of this free radical the activity of this free radical. So, that they are not unbound to do anything they want after the bacteria is killed you need to come back home suppose this is our hit man ok in the cellular system we have given a contract to kill the villains that is the bacteria. So, what they do they are checked by multiple enzymes the most important of that is glutathione peroxidase and glutathione reductase also superoxide dismutase.

So, mind it superoxide dismutase is a part of generation of reactive oxygen species it also tackles some reactive oxygen species by scavenging them, but the most important is this word glutathione and specially is in the reduced form and who helps in reducing glutathione the enzyme glutathione reductase. Now, the question is we will be seeing everything in our diagram very soon who actually helps in reducing glutathione for reduction we need a hydrogen ion right this hydrogen ion is actually supplied by NADPH. So, watch very carefully what is happening over here now we are trying to tackle the free radicals that are already in excess. So, NADPH by the action of NADPH oxidase was generating free radical one paradigm next is. So, again we are getting NADPH from glucose via the HMP shunt what is happening the hydrogen from NADPH helps in maintaining glutathione in its reduced form.

So, this is the oxidized form of glutathione and this is the reduced form of glutathione this is denoted by GSH two molecules of reduced glutathione G to GSH and when in oxidized form they actually dimerizes to form GSSG a disulfide bond is there. So, remember anywhere you are include listening to the word thio it means sulfur, sulfur is there ok you might be knowing that from your chemistry knowledge already. So, what happens? So, these glutathione peroxidase is a variety of peroxidase. So, what it will do?

It will convert the peroxides to harmless water right. So, where from we are getting peroxide superoxide dismutase can convert all free radicals the reactive oxygen species that O2 minus etcetera to H2O2 ok.

This you already saw in the previous mechanism H2O2 after being produced when we are killing bacteria myeloperoxidase act on that, but here glutathione peroxidase is acting to produce water which is absolutely harmless to the system right. So, what will happen if so, can you guess if this thing is jeopardized what will happen? See there are multiple source of free radicals. Free radicals are actually not only produced by NADPH oxidase there are other multiple cellular sources of free radical which are produced from either cellular aspiration at mitophonia oxidation there are oxidative drugs there are multiple sources. However, this is the only source or major source by which all the free radicals can be scavenged. So, if there is a deficiency of NADPH ok there will be less and less glutathione reduced glutathione there will be less and less scavenging of free radicals and this free radical will ultimately destroy all the membrane specially the RBC membrane in which there is a huge amount of HMP shunt going on ok.

So, this is an applied question right over and how NADPH can be deficient can you tell me glucose produces NADPH by the action of glucose 6 phosphate dehydrogenase we are talking about deficiency of NADPH. So, who is at fault? Definitely this if this enzyme is deficient the only problem we will be discussing it very soon. So, this all leads to an loss of integrity in RBC membrane. So, normally RBC membrane is maintained intact due to action of NADPH glutathione and glutathione reductase that I have already explained in the previous diagram. So, if those free radicals are left unchecked it will lead to loss of or destruction of RBC membrane.

So, in when we are looking at the beneficial role NADPH glutathione and glutathione reductase actually helps to maintain the integrity of RBC membrane since NADPH the major source is HMP shunt. So, the existence of HMP shunt is actually protecting the RBC or erythrocyte membrane from free radical damage ok. The next importance of NADPH mind it we are discussing the role of NADPH ok I hope you are there with me. So, NADPH is also important in prevention of methemoglobinemia right what is methemoglobin? Methemoglobin is the oxidized form of hemoglobin in remember in hemoglobin that is the pigment that is in RBC that is carrying oxygen is in ferrous form Fe 2 plus. It has got a tendency to be oxidized to ferric form Fe 3 plus.

If iron in hemoglobin is oxidized to ferric form hemoglobin is converted to methemoglobin and it loses all capacity of carrying oxygen ok. Methhemoglobin cannot carry oxygen and who helps to maintain the reduced state again NADPH. So, NADPH is required to keep the iron in ferrous state and to prevent methemoglobinemia. So, again if

there is deficiencies of NADPH there will be methemoglobinemia right. Next NADPH again one of the very important uses to help in detoxification of drugs.

You just need to know most of the drugs that you are consuming medicines ok they after their action they need to be rendered useless. It happens in liver in the microsomal oxidation system and it uses an enzyme system that is known as cytochrome P450 based oxidation system cytochrome. It is the iron containing pigment. So, cytochrome P450 based system is helping in detoxification of drugs in liver and who helps in the cytochrome P450 system again NADPH. So, if there is no NADPH there will be no detoxification of drug or there will be very reduced detoxification of drug using cytochrome P450 microsomal enzyme system and suddenly all the drugs will have more and more side effects ok.

Next NADPH is also important in eye to maintain the transparency of the lens ok. Just know this NADPH is very important in preserving the transfer transparency of the lens because if there is excess oxidation of the products that will lead to osmotic imbalance there will be water intake and all the refractive index of the lens will be blurred and then we will have something known as that that will be very problem vision will be blurred right. So, NADPH being present in the lens also helps to keep the essential lens components in reduced form and thus maintain the maintain the transparency of the lens. So, what happens over here? So, for production of NADPH we already learned the main important enzyme the key enzyme is glucose 6 phosphate dehydrogenase. After knowing all these thing that glucose 6 phosphate dehydrogenase is doing and after all the beneficial roles of NADPH you can easily guess what will happen if there is a deficiency of glucose 6 phosphate dehydrogenase.

So, even if you do not watch this video along you can pause this when you can actually make your mind and make just make few notes what will be the problem let the video run and you can tell you later where you write in predicting. So, let us see ok. First of all the G6P deficiency is a sex linked trait ok. It means what? It is always dormant in females because there are two X chromosome which is an X linked recessive disorder and only in male patients or male individuals where there is only X 1 X chromosome and 1 Y chromosome they can be expressed. So, all G6P deficient cases are males females are carriers of G6P deficiency fine minded males are patient and females are carriers they are patients or their cases right.

So, G decrease G6PD impairs the synthesis of NADPH in RBC that will lead to destruction of RBC membrane and that will lead to hemolysis right. This hemolysis is more and more pronounced when the body is all under some oxidative assault. For example, drugs like primaquin there is an anti malarial drug chloroquine and primaquin.

Primaquin is very essential for treatment of malaria for drugs like primaquin and few vegetables like fava beans ok. They contain some products which leads to increased oxidative cellular environment and when their oxidation is happening more and more oxidative drugs they produce more and more free radicals and we need the action of NADPH glutathione reductase etcetera to scavenge those free radicals.

So, a normal person you and me who does not have any problem will can easily intake primaquin fava beans and we are fine because all the NADPH are scavenging the free radicals. What if this NADPH is deficient because of deficiency of G6PD then suddenly maybe normally we are going fine with some amount of NADPH that is being present ok from malic enzyme, but as soon as there is an oxidative assault there will be profuse hemolysis because there is no one to scavenge all the free radicals and they will destroy the RBC membrane right. So, if there is an excess production of free radicals. So, we need more and more reduced glutathione we need more and more NADPH, but if this is deficient there will be no NADPH there will be no reduced glutathione and ultimately these free radicals will destroy the RBC membrane they will membrane will be totally destroyed and that will lead to a disease known as hemolysis that is restriction of red blood corpuscles. Now, this after this hemolysis occurs what happens you know the heme first of all hemoglobin is goes out of the cell and what happens this hemoglobin can be no longer maintained in the reduced form because I told you NADPH is also essential in in preventing the conversion of hemoglobin to meth hemoglobin.

So, there will also be meth hemoglobin in here. So, when observed on the microscope this hemoglobin molecules cross link with each other to form something known as Heinz body again very important MCQ question. So, membranes are and these Heinz body also plays a role in damaging the RBC membrane. So, RBC membrane by Heinz bodies and reactive oxygen species due to deficiency of NADPH which is due to the efficiency of jucilosic phosphate dehydrogenase leads to hemolytic anemia all the RBCs will be gone hemoglobin concentration will fall anemia will happen and it is a condition known as hemolytic anemia ok.

So, G6PD deficiency and malaria ok. Now, G6PD deficiency is actually associated with resistance to malaria this is a protective mechanism that is found in African population. So, what actually happened over here with adaptation the malarial parasite that is invading the red blood cell actually needs reduced glutathione for survival right. And the African population some areas of African population has been seen they they are born with glucose 6 phosphate dehydrogenase deficiency right. And those areas are actually prone to tropical diseases like malaria. So, imagine the situation what happened if they are contracting if they are having malaria the disease if they are giving prima quin hemolysis or hemolytic anemia will occur and the patients will die.

Naturally I mean luckily they have been protected by evolution in such a way that the malarial parasite itself needs reduced glutathione right to survive. So, in those patients where glucose 6 phosphate dehydrogenase deficiency is there, there is no NADPH and there is reduced glutathione is also less. So, the malarial parasite dies in the first place. So, they are naturally protected from malaria. So, this is an example of a natural adaptation where an enzyme deficiency in a specific geographical region is protecting the population from any disease the disease being malaria.

Next thiamine deficiency did you remember the enzyme trans ketolase the enzyme trans ketolase was catalyzing transfer of carbon atoms from 5 carbon to 3 carbon into the formation of you remember in HMP shunt if you do not please there are 2 trans ketolase reaction. You should remember that you should pause the video look at the previous class look at the pathway of HMP shunt and then come back here. Over there it was mentioned the thiamine is actually the cofactor of trans ketolase enzyme in the form of TPP or thiamine pyrophosphate ok. So, it is so much dependent that actually the deficiency of vitamin B 1 or thiamine is checked by estimating the trans ketolase activity. So, if the trans ketolase activity is down the vitamin B 1 deficiency is there and this is actually manifested by a disease that is known as Wernicke's Korsakoff's syndrome.

In many text book it is if the disease can be split I mean the milder phase is known as Wernicke's NK phallopathy and the severe phase is known as Korsakoff's psychosis, but in many text book they are clubbed together. So, Wernicke's or Korsakoff's you mind that this is vitamin B 1 deficiency thiamine testing by trans ketolase activity. What are the features? The features are loss of memory, mental disorder and partial paralysis. So, these are the significance of HMP shunt that have been discussed. So, reductive biosynthesis of fatty acid cholesterol and steroid hormone for next set of classes free radical scavenging maintaining the integrity of RBC membrane, prevention of methemoglobin formation, detoxification by hydroxylation maintaining the transparency of lens, bactericidal activity of macrophages this will again will be discussed later ok or even in live classes for that matter.

So, to produce and the importance of the non oxidative phase is to produce DNA and RNA with the help of pentoses and clinically what you need to remember is glucose 6 phosphate dehydrogenase deficiency that actually leads to so many problems ok. So, the whole problem is due to this G 6 B deficiency and this also leads to drug induced hemolytic anemia, methemoglobinemia as well as thiamine deficiency that leads to reduced trans ketolase activity. So, the key points in this class where the whole combined two classes is NADPH is produced as well as pentoses are produced from HMP shunt.

The first reaction that is catalyzed by glucose 6 phosphate dehydrogenase is the rate limiting step and it is inhibited by NADPH as an allosteric inhibitor. The oxidative phase is controlled by the level of NADPH and non-oxidative phase is controlled by requirement of pentoses and insulin induced glucose 6 phosphate dehydrogenase and therefore, increase the overall pathway ok.

So, these are my references and thank you all for your attention.