Human Physiology
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Lecture – 28
Excretory system: Nephron - Part 1

With this I come to the end of what lecture? I come to the end of 7 lecture and I will move on to the, okay we are back to kidney now. What we did so far was this erythropoietin, skip it, aside for the time being now. So far we have been focusing on Bowman's capsule and essentially the events that happen there, the flow of blood in the flow of blood, the ultrafiltration and the different factors that we have been focused on Bowman's capsule. But what you get there is the filtrate, it is not urine, okay. For the production of urine, it has to be processed and for that it has to go all the way down in the, so Bowman's capsule, proximal convoluted tubule, here is the Bowman's capsule, okay and here is the filtrate and it will go here, then it will go into the, okay let us see. This is the Bowman's capsule, proximal convoluted tubule, okay and then thick segment and then from here comes a loop of Henle, follow the story, loop of Henle thin, thin segment, thin segment descending, thin segment ascending, thick segment ascending, we will call 1, 2, 3 parts, thin descending, thin ascending and thick ascending, did you get the three names? Thin, thin, thick, okay, thin, descending, ascending, thick, okay.

We will call, put together, we will call it as what? Loop of Henle and as the urine, as the pre-urine flows throughout the entire and eventually beyond, it is being processed and leading to the formation of final urine, that is ready for discharge. So, what we are going to do in today's lecture is try to focus on how the, how the urine is processed. Now what I will do is, I will take, okay you know this image, this is the Bowman's capsule and from here is the proximal convoluted tubule. You can see, see the proximal convoluted tubule is convoluted, okay it goes up and down, why do you think it is, what may be the rationale in making the tube convoluted? Perfect, perfect, because you need to absorb a lot of things, you need to, so it is a good idea to make it this way.

So, what I will do now, I will take a tiny part of the proximal convoluted tube somewhere here and I will blow it up so as to show you the different components. And I have, what I have here is the, is the tubular lumen, so this is the filtrate and the filtrate is going and as it is going it is interacting with the cells of the tubule and the cells of the tubule are single celled, it is in epithelial layer, it is a tube with single layer of cells, okay. And here you have, so we will call them as the tubular epithelial cells and there is one tubular and two and three and at the, and they are connected to one another by means of what a special type of proteins which make the, which make what you call as a tight junction, what do you call it as? Tight. You call it as the, call it as a tight junction and on the other side you have the interstitial fluid, here interstitial fluid and they have, and we have a capillary. Now what you need to do is, you

have to remember, you have to remember this picture because this picture is going to repeat all over as we go along the length of the tube, we are going along the length of the tube and the function of the length of the tube keeps on changing.

But it will always be the three components will be there, what will you have? The lumen through which the filtrate is passing and it will be lined by the cells which we call as the tubular epithelial cell, the interstitial fluid and there will be peritubular capillary. What do you think will be the pressure in that capillary there? We did it yesterday three, four times. What do you think will be the pressure in this? Yeah, you are right. 15. 15, 10, 15, okay, some place it will, it is very low, it is very low, okay, all right.

Now, more couple of terms to be introduced to you, this phase of the cell, which is directly coming in contact with the filtrate will also call as the apical face, what do you call it as? Apical. Apical and the other one I will call as the basal and these two face I will call as, actually there are four, are you with me? If I presume it to be a cube, which it may not be, presumably then there will be six faces. The other four faces I will call as basolateral, I can see basolateral, basolateral, lateral, basal and apical. Every face has its own importance, please remember, apical, basal, basolateral, are we good so far? Good. This just gives us a summary of the treatment of different molecules and ions that happens as the filtrate travels along the Bowman's capsule enters into the proximal convoluted tubule so far.

Here the proximal convoluted tubule ends and then we have the descending limb of the loop of thin ascending and then we have this is all, this is all and then okay, so let us see, what does this K+ indicate, Na+ indicate, amino acid, what does it indicate? They are being absorbed. They are being absorbed. So, there is K+, Na+, amino acid, glucose, water, not much action is happening in this part of the tubule, but then suddenly when it becomes thick, Na+ lots of Na+ is being absorbed. Here K+, look at the arrow, K+ is in opposite direction, K+ is being secreted, H ions are being secreted and finally water is being absorbed and this gives us, if you take transverse section at different levels, it tells us how the proximal tubular loop of Henle is extremely thin if you compare the two, are you with me? And then the collecting tubule will be somewhere here and the distal convoluted tubule which will be distal convoluted tube is here and these will help us to understand as to how it works, okay. This is essentially the same figure.

Can we immediately identify filtration? So, the fluid is going this way, are you with me so far? Okay. So, if that is the thing, then this villi that you see, this microvilli that you see is the apical side or basal side? Apical side. Apical side, good, good. And then here you have the entire cell and there is the interstitial fluid and there is a capillary. In this image, Guyton tries to tell us as to what are the different ways by which the different elements or different ions can be absorbed.

Number one, either the okay, now here is a cell and somewhere there is a tight junction. Now the tight junction immediately gives us an concept that its tight and nothing should go, but we are wrong because although it is tight junction, the water molecule is so small that they can go, sodium+ ion is so small that it can go, larger than that cannot go, glucose cannot go. Are you getting my argument? Author is trying to define us what tight junction can and cannot do, tight junction will allow water to go, sodium ions to go, yes, chloride ions to go, yes, but okay. Then the some of the substance can directly enter into the cell, into the cytosol of the cell and then they can grow and then the author tells us about the ATP molecule that makes heart and soul of the whole system.

Okay. What are we talking about? Another 15 minutes we are focused on the proximal convoluted tubule. Can you see the diagram there? Can you see the Bowman's capsule there? Can you see the convoluted proximal tubule there? Okay, Guyton has highlighted it. Why? Because now we are just focused where? Where? Proximal convoluted tubule and that is where we are going to say what is going to happen and here is the summary, okay. What we are going to take out of that is sodium, sodium chloride, bicarbonate, potassium, water, glucose, amino acids and we are going to secrete some hydrogen ions and organic acid. This gives us a little now.

Can you appreciate this? What are we looking at? Transmission electron photomicrograph of these tubule cells, okay, section through the proximal convoluted tubule, okay and can you, this is the transverse and this is the scanning electron photomicrograph. Can you see the villi there? Huge number. This actually should remind you of the brush border when we refer to in the intestine. Hello, are you with me? In the intestine we have the brush border, exactly brush border, okay and the logic is again same - the cell wants to increase enormous amount of surface area, okay because there are huge amount of proteins there which are selectively doing a lot of function of importing lot of things from the lumen to the interstitial fluid. I will talk about it later and this gives us a scanning.

So these are all cells, this is the basal side and these are the actual cells and this shows the cells in and, and, and. Within the cell which is very, very prominent cell organelle that you can visualize. Yeah, you are right, right. Mitochondria, how many mitochondria can you see? Huge amount of mitochondria. It is a very energy expensive one because you have to ensure that a lot of ions etc.

transported against the gradient, okay. So you are going to need ATP and therefore you need to, you have, you have to have a large number of, this is the basal side, what am I showing you? Again look at this part. If you follow it very carefully you will see that there is a descending limb, there is ascending limb and there you will actually find that there is a proximal convoluted tubule and a distal convoluted tubule and they are almost entangled with one another. They are not really. So if I take a transfer section, hello, if I take a transfer section here I am going to get proximal, proximal distal.

What is it, what is author trying to tell us? Hello, means okay, Bowman's capsule so far the tubule goes down and then proximal convoluted tubule there goes hundred times up and down and goes down, are you okay so far? Then comes up and then distal convoluted tubule and the two come very close to one another and that facilitates transfer from here into the interstitial and then into the distal convoluted tubule. If I take a transverse section there what am I going to find? The proximal convoluted tubule transverse section and the distal convoluted tubule and transverse section but they are very, very close to one another and on the basal side okay. Yeah, it is, it is, it is because the, it is very different okay. I will come to that, we will come to that. Right now we are focused on proximal, I will tell you the way these cellular, these cells handle the fluid that is flowing is very different, very different.

We are still with proximal convoluted tubule okay. So we have a section okay. So we take kidney of rat, then fix it, fix it, cut frozen sections in a cryotome okay at minus 30 degrees okay and take the sections. Now this time we treat the sections, earlier time we treated the sections with antibodies against erythropoietin. Now I am going to apply on the sections the antibodies against, against sodium potassium pump ATPase, ATPase, we have done it in earlier semesters.

ATPase, hello, yes or no? Yes. Great, great. So we take the antibodies against that and we find that these are the, these are the tubular sections okay and when I cut a tubule in the section I find that this bluest part represents the lumen, this part represents the cells and on the basal side of the cells I get the immune-reaction. Are you with me there? Look at, so cell is here, here, here, here, here, here, here, the cells are facing this way that is the apical side, this is the basal side and these are all the basal. So these are several cells and on the basal side I get the immune-reaction which means ATP molecule is located in which membrane of the proximal convoluted tubule? Basal side.

Basal side, what side? Basal side. Please remember that and now we are ready to see this cartoon. Instantly you know what I am talking about, what is this, what does this represent? The lumen, the lumen, what is the filtrate is going down? We are in the proximal convoluted tubule and the freshly arrived filtrate is going that way and then these are the cells, these are the tight junctions, this is the interstitial fluid and this is the capillary there. Done so far? I find that in the apical membrane here, in the apical membrane, remember, you see, remember that electron photomicrograph slide, remember those microvilli, okay, see, the hundreds and hundreds of protein molecules, okay, which serve as a symport system, symport system which will take along with sodium, it will take glucose, okay, so far. Now, why is sodium going inside? Point number 1, okay, let us use, let us ask a common sense question.

In this fluid, as it is filtered in the Bowman's capsule and flowing along the proximal convoluted tubule, how much do you think is sodium ion concentration in that filtrate? 140. Absolutely correct, I am so happy, how much it is? 140. 140, that is in the blood, it is just coming in the blood and everything and it is going to be, it is going to be in the same

concentration, it cannot be more, it cannot be less, okay. Now, here, so the concentration here of sodium is 142 and in here, how much is this concentration of sodium ions in the cell? 10. 10, okay and then, and then here it is the, in the lumen, it is about, about the, if you actually put an electrode there, it is about minus 3 millivolts, inside is about minus 70 millivolts, hello, okay and the sodium is 142 and the sodium is 10 and then you have a carrier there, what is going to happen? With the help of the carrier, the sodium will go in, okay, this is minus 3, inside is minus 70, outside is 142, inside is about 10, 12, whatever it is, okay.

So, the sodium will drive the transport and along with that, the glucose will get a transport – symport system. So, in the cell now, you have, you have more sodium, okay. But at the same time, at the same time, look at the basal side, the side which is towards the capillary, what do we have? What do we have? We have already seen that in the previous slide, we have the demonstration from the immunochemistry protocol, are you okay there? Okay, so, so the pump is there, okay and to make sure that the pump, to make sure that every time pump goes through a cycle, pump goes through a cycle, means 3 sodium ions to the outside and 2 potassium ions to the inside, every time it goes through a cycle, it will use one molecule of ATP, it will keep on doing that and as a result of that, as a result of that, the sodium ions in this will be less and as a result of that, there is a great drive for the sodium ions in this lumen to be taken into the cell. So, there is concentration gradient, there is electrochemical gradient and there is a carrier, so it will go. Just as there is a carrier for sodium and glucose, okay, I will tell you in the next slide, it is called as SGLT, you can immediately make out, S stands for what? GL stands for what? T stands for what? Transport.

Okay, it comes in that protein system, comes into 1, 2, 3, 4 forms, but do not bother about SGLT protein, SGLT protein, this protein is going to come very repeatedly come back to us, SGLT protein, okay. It is a carrier system, it is driven by sodium ion, okay, it is a secondary transport, it depends on the primary transport, the primary transport is because of the pump which is on the basal side, we are good so far, alright. Just as there is this protein system that accounts for the symport of sodium driving glucose, we have other protein systems on a very similar scale, which will along with sodium will drive what? Amino acids, some ions, some vitamins, etc, lot of things. We are just taking one example of sodium chloride, but there are other systems also and therefore that huge number of what we like, okay, on which the protein systems are sitting. Okay, now if the sodium has gone from here, okay, and then from here it travels and because there is more sodium here, it just goes passively, there is nothing to drive sodium into the capillary, but it will be in equilibrium, whatever goes here, it will be in the equilibrium and some of the sodium will get into the capillary.

Now, it is not at all a matter of surprise that if sodium goes, sodium has gone so chloride will go, okay or not? It has charge, okay, it will just flow - if sodium has gone and if sodium has gone and chloride has also gone, which means osmolality has gone. Hello, okay, and if the osmolality has gone, what will go now? Water, what will go now? Water, so in this way, so this is how the water goes, this is how the chloride goes, okay, this is how the urea goes, how urea goes, I will explain in some details. So, the chloride ions take a paracellular

transport, which means, so chloride ions being small, how are they going? They are going via the? We are still talking of the same story, but we are going into some interesting details as to we are talking about the proteins now. Look at this image. Whenever we look at the first thing we get our bearing, okay, I should know what is north, south, east, west.

In this particular case, what do I look for? Where is the lumen? Hello, okay, so this is the lumen, I know that the urine is flowing this way, okay, and I know that this is the interstitial fluid and blood means my capillary is somewhere there, okay. And I find that in the on the apical side, which is so I have already seen that what is the what does it stand for? What kind of SGLT is there? I told you there are three are there, okay. In the kidney, what kind of SGLT do we find? Two kinds of protein system, you know, proteins are notorious for coming in different forms. So, this SGLT takes sodium and glucose now, this so now we have a lot of glucose, a lot of glucose there and we want to make sure that we want to absorb every glucose molecule is there in the now this glucose is now needs to be taken into the capillary, okay, but glucose molecule on its own cannot really cross the membrane. Therefore, the plasma membrane on the basal side is equipped with another protein system, which I will call as tell me what? GLUT2, can you tell me what GLUT or GLUT2 stands for? It is a glucose transporter, what is it? It is a protein system which I call as a glucose transporter and that glucose transport but please remember, glucose transporter has no sense of directionality means what? If it is more here, it will take there if it is more here, it will take there, get it, it has no directionality that protein but as it happens, as it happens here, there is more because of this SGLT system, there is more glucose here, there is more glucose naturally flow from here to the outside and from there if it builds here, then it will pass on into the into the capillary.

What are we doing? We are absorbing, okay, we are absorbing what? Sodium, chloride, water and glucose and on similar mechanisms we are, we are, where are we? Proximal convoluted tubule, where are we? Yeah, we are still in proximal convoluted tubule. Yes. Are these GLUT2 channels insulin independent of the, or as a matter of fact are any glucose channels like facilitate glucose from intracellular to extracellular environment, do they need insulin that they need for extracellular to intracellular task? Okay, as I said, a GLUT will take you from anywhere to anywhere, more to less, higher, higher concentration to lower concentration. I will focus on two types of cells, one is a neuron and one is a muscle cell or an adipose cell, it is a muscle cell is good enough for my work. Now, you are starving, no food for last 6 hours.

As a result of that, the blood sugar level has gone down, it has gone to about 80 mg and when the blood sugar level has gone down to 80 mg, your beta cells - the secretion of beta cells has almost come to negligible, which means no, no insulin is being secreted and as a result of that in the, in the, okay, okay, good, good. Now, you have taken food, as a result of that blood sugar level has gone up, the blood sugar has itself acted on the sensors which are located in the plasma membrane of the beta cells of the islets of Langerhans, okay and as a result of that insulin will be secreted. How that will happen that I will tell you when I talk

about insulin, insulin will secreted. Now, when insulin will go into the blood, it will act on the insulin receptor which is sitting on the plasma membrane of the skeletal muscle cell, okay. Once that happens, it will take a signal from the plasma membrane inside the cell and inside the cell there are, there are inside the cell muscle cell there are vesicles and those vesicles in the plasma membrane will have GLUTs.

Where is it? In the, within the cell, okay. Under the influence of information that is coming from the insulin, the vesicles will now be transported, they will go and be assimilated in the plasma membrane so that now the GLUT that was in the vesicles is now exposed to the outside and now the blood glucose level that has gone up because you had lunch, now it will now that glucose will transfer via the GLUTS now which is in the plasma membrane will go in. It will remain there for 15 minutes after that, after that that GLUT will again be internalized. Are you with me? Therefore, the transport of glucose across the plasma membrane of a muscle cell and of the adipose tissue is insulin dependent.

Good? Go back to brain. Is everybody, it is very, I will actually I would have come to this story eventually but your neurons need glucose every second. Can you afford to let them, can you afford to let the system remain at the mercy of the GLUTs which is in the vesicles, it will go there, it will lodge and then no, no, no, no, no, no, no. The brain cells have GLUTs which are permanently sitting there, constitutionally there, okay. Therefore, as far as our brain is concerned, the neurons are concerned, different types of GLUTs are constitutionally sitting there, so they do not worry, you take glucose, the GLUTs are there, they will take the glucose from the blood into the interstitial fluid, into the neurons, it will be metabolized. This is complete answer to your question? Great.

So, look at, any other question? Okay. So, look at this, what am I showing you? Just appreciate the beauty of SGLT family. We are looking at what? One of them, one of them, okay. Where is it sitting on the apical side or basal side of those cells? Apical side. If it is apical side of the proximal, this is SGLT of what kind? It is SGLT2, you are right, it is SGLT2. And look at the protein, how many times it goes up and down? Fourteen times, fourteen times, a huge protein molecule, huge protein molecule.

It does what, symport of what and what? Sodium and glucose, driven by the power comes from sodium, okay, and then it drives. So, this is a, the author tells us SGLT1 is mainly in the intestine, heart and kidney, okay. So, the food that we take, carbohydrate digested into glucose from the lumen of the intestine is taken up into the, into the, into the, into the capillaries of the intestine. So, as it travels along the, the, the, the brush border of the intestinal cell, what kind of SGLT will be located there? One, okay, one kind. And then, okay, and then you can read more about it.

We again take the section through the proximal convoluted tubule, sections. We will get the same sections, we already have, we already have the sections. This time you stain them with antibodies against SGLT, okay. And therefore, on the, so, so, so this is actually is the lumen of the proximal convoluted tubule and coming right in contact with the lumen is the plasma membrane of the epithelial cells, okay, and that membrane is, is known to have what protein? SGLT2 and you, since you had antibodies against SGLT2, you are, you have labeled, so what you are looking at is SGLT1 here, 2, 0, whatever you, okay, in this. So, so, so this is the kind of immunoreaction you will, you will get.

Now that we know about SGLT1 and 2, let us also take time off and try to find out about, about, about, about what? About what? You tell me, you tell me. It is written there. What is it? GLUTs. GLUTs, okay. 1, 2, 3, 4, at least 4 different types of GLUTs are there, okay.

So, we are talking about glucose transport, a wide group of membrane proteins that facilitate the transport of glucose over the plasma membrane. GLUT2 has bi-direction transporter. We have already made the time, point, transfer, allowing glucose to flow in two directions. It is expressed by the, it is expressed by renal tubular cells. So, in proximal convoluted tubule, what kind of GLUT 2 are you going to get? Here it is written, it is written.

What, what kind of glut is going to get? 2. It is going to be in the, it is going to be in the apical side or the basal side? Basal side. It is going to, it is going to, good, good, it is going to on the basal side, you are right. This is just a reminder as to, we have seen that on the basal side, we have this pump, this protein system, sodium potassium ATPase. This, I just put this slide to give you an overview of what are the different pumps that will, you will encounter during the entire tubule. And the one that we have already seen, look at the one, there are four points I have, I have, I have summarized for you.

What is number 1? What is number A there, please? Sodium potassium ATPase. Sodium potassium ATPase, means these are all pumps, they are going to, they are going to work uphill against the, against the gradient, maybe concentration electrical, whatever against the gradient and they are going to use ATP. And the second one that you are going to have is what? Hydrogen-ATPase, hydrogen ATPase. Where else do you think in your body you should get an abundance of hydrogen ATPase? Yeah, you are right.

You are right. You know something? Our stomach secretes huge amount of HCl. So, stomach has proton pump. What is a proton pump? Hydrogen-

Good. Hydrogen, hydrogen potassium ATPase. Just as there is sodium potassium, hydrogen and there is calcium ATPase. So, just remember that, that, that, that there are four different

types of pumps. They are all going to use energy and they are all going to bring about the transport against the.