Kidney Complications and Lysosome Disorders

Ozlem Goker-Alpan:

Hello everyone. I am the host of the CME Series on Lysosomal Disorders. I am Ozlem Goker-Alpan, and I would like to thank everyone attending today to our last series in this year. We're going to be talking about kidney involvement in lysosomal disorders. As a brief intro, you may be aware that actually there is involvement of the kidneys in most lysosomal disorders this and that way. There are rare case reports that is with the kidneys getting affected, either patients presenting with proteinuria or other signs or symptoms of kidney issues. So why the kidneys are so important for lysosomal dysfunction? Because the lysosomes actually are primarily responsible in kidneys for recycling of low molecular weight proteins that cross the glomerular filtration and also regulation of water absorption by principal cells and the electrolyte homeostasis.

So in healthy glomerular cells there are various expression of lysosomal membrane proteins that suggest the existence of subsets of lysosome or endosomal vesicles with podocyte mesangial cells and glomerular endothelial cells. And we'll be discussing for example, why podocytes are invariably involved in Fabry disease and some other lysosomal disorders. Similarly, in this figure what you're seeing is LIMP-2 positive vesicles are also predominantly defined in mesangial glomerular endothelial cells. Why this is important? Also the ones that are familiar with LIMP-2, is the transport protein for glucocerebrosidase. But there is a disorder that involves the variance in the gene encoding for LIMP-2 that is responsible for a disease that is called myoclonic epilepsy and renal failure syndrome.

I am told that my video has not been started. Okay. All right. So basically we said that the lysosomal disorders, they do primarily affect glomerular cell types differentially due to potentially different capacity in their autophagy or lysosomal functions. So this is a whole list of... Actually, it's a partial list of the lysosomal disorders that can present with renal involvement. However, the two primary lysosomal disorders, when we think about lysosomal diseases, first is cystinosis where a transport lysosomal protein involved actually presents with tubular dysfunction or Fanconi syndrome. And the other prototype disease that leads to end stage renal disease is Fabry disease.

So a nephrologist can see either patients with cystinosis or patients with Fabry disease without them realizing that they do have a lysosomal disorder. So a nephrologist can be the diagnostician in this case, so they can take a role in early detection and screening. So that is why it is of utmost importance for a nephrologist to be acknowledged of at least these two primary disorders that presents with kidney involvement. And they are also the ones that could be performing the kidney biopsies and also interpreting them. And in our patient cohort, almost 20 to 30% of patients that come with a kidney biopsy result with the presence of zebra bodies, in the case of Fabry disease. So they get diagnosed retrospectively with Fabry disease despite that they did have the symptomatology that started probably earlier years, in case of patients with classic Fabry disease.

And also they would be responsible from interpretation of the lab findings, also the genetic diagnosis. We know that the nephrologists work with closely the geneticists, and then geneticists can lead them to understand what the genetic diagnosis means. But they would be the ones that would be interpreting the progression of kidney disease and other findings in the laboratory diagnosis and the follow-up. And also, they would be mainly the ones that would make a differential diagnosis when there are complicated cases, such that we have a patient with Fabry disease who has also IgA nephropathy. Basically these complicated patients require a nephrologist's attention for further management and diagnosis, and also similarly that they could be the consultants in the case of patients who are recently diagnosed with Fabry disease and we want them to be evaluated by a nephrologist to understand the

status in their renal involvement, and further, that they do have a role in patient education and counseling.

So they do also have primary roles for comprehensive care and treatment approaches in patients with lysosomal disorders, in case of Fabry disease. So they are the ones that manage kidney-related symptoms and complications, like we had a patient who came with hyperkalemia recently, and the nephrologist was the one actually who introduced the proper treatment for that. And similarly, the patients who are requiring renal replacement therapy, the nephrologists are the primary physicians that would decide their management and care. And obviously, they are the ones also that collaborate with the research community to advance our understanding of the lysosomal disorders. So I would like to take this opportunity to introduce Dr. David Warnock, who is the esteemed professor emeritus at the University of Alabama at Birmingham. David Warnock has made significant contributions to nephrology and medicine in the field of lysosomal diseases, especially Fabry disease. He has a career spanning for several decades, and his work encompasses groundbreaking research, particularly in the field of Fabry disease.

His academic journey is marked by numerous awards and honors, reflecting a deep commitment to advancing medical science and education. And as a recognized leader in the field of lysosomal disorders and Fabry disease, Dr. Warnock's expertise and insights to the kidney involvement in this unique disorder and its complex interactions with the system's biology have been invaluable to both the scientific community and also understanding the patient care.

Now it's my pleasure to introduce Dr. David Warnock. Thank you. Today we welcome Dr. David Warnock for a fireside chat. David, so as an opening question, could you please talk briefly about the kidney involvement and injury that occurs in Fabry disease? Actually, before you start that, I have a burning question. So why podocytes are so vulnerable with the involvement of the lysosomal system?

David Warnock:

Well, some would maintain that the podocyte is the heart, the soul of the kidney, and I do think it's very important. It's part of the whole filtration apparatus. As you well know, the kidney starts, the individual unit is called the glomerulus. It's really a filtering unit, and the filtration membrane itself is maintained and generated by the podocytes. So the podocytes are very important. But with respect to our interest in lysosomal storage diseases and the treatment of those diseases, they play a very important role, because unlike other critical cellular elements, they are not directly in contact with the bloodstream. They're very similar to the cardiomyocyte in that respect. Both are terminally differentiated, both, if they are lost through injury, apoptosis, whatever the process is, they are not replaced, at least not in our current understanding. Because they are not directly bathed in the bloodstream itself, when we inject intravenous enzyme replacement therapy, they're not immediately accessible to that enzyme.

The enzyme has to traverse the filtration barrier itself to get to the podocyte side of things. Same story with the cardiomyocytes. Now that opens the question, begs the question, what about oral systemic therapies, chaperones or perhaps the substrate reduction therapies? Those do have a better distribution, if you will, to all cellular elements. Some even penetrate the blood-brain barrier, as you well understand. But I think in the context of our conversation today, there is, to my mind, such important and exciting new information in the enzyme replacement therapy field, I thought we could spend some time talking about this.

Ozlem Goker-Alpan:

Absolutely, we will. Let's just a little bit touch base about the events that occur in Fabry disease that actually determines the disease onset and the progression and the stage that the patient in, because this is also important about the treatment outcomes for any patient. Could you just briefly mention about that before we delve into the enzyme replacement therapies?

David Warnock:

Indeed, if I could, I'd like to show a slide. I have very few slides I want to share today, but this is important, and it shows the progression in terms of first cellular and then organ damage with respect to Fabry disease. So Ozlem, when this slide is called Fabry Disease Accumulation, Cellular Injury, Compromise Function, and Organ Failure, and it represents each of the four stages of involvement. Now, as you know, I'm a nephrologist. I care and treat patients with kidney disease. I'm a nephrologist with a focus on adults with kidney disease, and so this slide is in the context of the kidney, but you could make a very similar slide for the heart, for the brain, or whatever organ system you want to discuss. The important point is that there is accumulation of the substrate that the enzyme is not properly processing, and that accumulation is in the lysosomal compartment.

These are lysosomal content enzymes, but in addition, we know that with injury, then there's some leakage into the cytoplasmic and other organelles and there's a processing, and it's a fascinating cell biology story that we could spend an hour talking about. But in the context of our discussion today, to go from cellular deposits, which even in the placenta had been described, this starts very early on, we start to see cellular injury occurring consequent to the organ accumulation, the cellular accumulation of these deposits. Now, that could be from the deposits themselves or toxic metabolites of those deposits. Again, another very interesting question, which I know you are very personally familiar with. We move from the phase of cellular injury now to organ damage, and that process starts with fibrosis. In the context of the kidneys themself, we start to see podocytes injured in the second phase. But in the organ damage phase, we start losing podocytes.

In fact, some have spent a lot of effort measuring the number of podocytes in the urine, podocyturia. Nephrologists love to have fancy words to describe simple concepts, you know that. But anyway, once we get to this stage three organ damage phase, then we start to see protein in the urine. We start to see a decrease in the filtration rate related to a rise in the serum creatinine. That's how we estimate this. Well, that damage is already occurred. Our focus should be in the cellular injury phase to try to intervene before the damage is irreversible.

Ozlem Goker-Alpan:

Even in pediatric patients, the podocyturia has been described, so which means that actually fibrosis starts much earlier, and these events do not take place consequentially, but it's kind of, it's in the three-dimensional structure, probably occurring all at once. Is that right? Or what's your opinion about that?

David Warnock:

Absolutely. I've had to change my tune with our fellows and residents, because I used to say, "Well, effacement of the podocyte, which is a sign of its injury, is a consequence of proteinuria." We now understand that effacement can occur before actual proteinuria, so we can demonstrate the cellular injury phase by biopsy when everything else seems normal. So we've used that as a tool to help us decide when to start treatment.

Ozlem Goker-Alpan:

Okay, so let's actually talk into the treatment right now. So the standard of care for a patient with Fabry disease is still the enzyme replacement therapy. And so what has been our experience, since you are actually been in one of the forefront of the different ERTs, the traditional enzyme replacement therapy that has been used actually for more than a decade now, what has been our experience about the dosing, the frequency, and obviously the efficacy that's contingent upon when we start the treatment?

David Warnock:

Ozlem, one of the wonderful things about this rare disease space that we work in is that a single patient tells a story, and we can learn a lot from the single patient. I well remember one of our first patients saying to me, "Doc, don't tell me it's one in a million. I've got it. For me, it's a hundred percent." And I think that message is a very important take-home message, that every patient has a story to tell and we learn something from these patients. Now, to that end, I want to show you a slide showing the course, actually, the first patient that we treated in our clinic at the University of Alabama in Birmingham. And we'll show the slide now. This patient at the time was in his 40s, had a family history of Fabry disease.

An older brother who in fact was already at the NIH with our friend Rapha Schiffman being treated with an experimental form, the alpha-galactosidase alpha. And this patient came to me, was referred by his primary care doctor because he had protein in the urine. So I saw this patient in 2001, and in the interval I controlled his proteinuria doing the things that kidney doctors do. We used a low dose of lisinopril, an angiotensin converting enzyme inhibitor, and a very common drug that I know you are very familiar with. And I was able to control this proteinuria quite readily. In fact, most patients with Fabry disease have relatively low blood pressures, and it's very easy to control both their blood pressure as well as their proteinuria. However, in some examples, and we'll talk about those in a bit, in some examples, the proteinuria is really persistent. And trying to push the ACE inhibitor or similar drugs only causes the patient to have bad side effects.

The blood pressure drops, and that's a problem, a major problem. Well, anyway, you'll see on this graph, and what's plotted here is the estimated kidney function rate. For our discussion, a hundred is normal, don't worry about the units. But you saw the patient came to us with 2.3 grams of protein in the urine, high serum creatinines. The estimated GFR was falling, falling, falling. That patient said to me that he wanted to go to the NIH to see Dr. Schiffman to enroll in the experimental program, and I said, "You should do that. I don't have access to the experimental drugs at this point." I hadn't yet started on my Fabry voyage. So we started the treatment with the agalsidase alpha at the NIH, and I continued seeing him as a patient in my renal clinic measuring his creatinine and his proteinuria and so forth.

And despite the best efforts on Dr. Schiffman's behalf and on our behalf, he continued to lose kidney function at a rate that was very, very alarming. The rate was about 13, minus 13 ml per minute per meter squared per year. That's an alarming rate of loss of kidney function. That's the sort of thing one would see in a patient with severe diabetic kidney disease. Well, Dr. Schiffman and I actually had a very interesting conversation the first time we met face-to-face and decided that the experimental drug of course was very important, but we should continue with the standard of care for all patients with kidney disease. And at that point I had become involved in the phase three studies with the alpha-galactosidase beta and was familiar with that and knew that that offered approximately five times greater amount of enzyme based on its dosing than did the experimental form under investigation at the NIH.

Well, the patient said, "Well, how do you know this will help me?" It turns out a very interesting story. He was a lab technician and understood linear regression analysis, and said, "I've taken all my creatinine values and my estimated GFR and plotted it against time, and at this rate I'm going to be on dialysis before I'm 50 years old. What can you do about this?" I said, "Well, there is a new medication that has been successfully tested, and I believe is about to be approved for use in patients with Fabry disease in

the US." This was in the early part of 2023, and as you well remember, and if I get these dates right, I think April 25th, 2023, the alpha-galactosidase beta was approved for commercial use in the US, and this patient in fact received his first infusion in May. Now, he asked me this question, which I think is a very important question and one of my take home messages here for our chat.

He said, "How do you know this might help me? How do you know?" And I said, "Well, I don't know, but I do know that the clearance of these deposits is very much dose dependent. And if we do a kidney biopsy and there are still deposits in the blood vessel cells that line the kidney capillaries and in the podocytes, the elements we talked about already, then in my opinion, there would be a rationale for switching your dosing to a higher amount of medication." And so we did a biopsy, we had that analyzed, and indeed there were persisting deposits in the blood vessel lining cells within the glomerulus itself. Now the normal capillaries, the routine blood vessels, had been cleared with the lower dose of enzyme replacement therapy, yet the kidney blood vessel cells of the filtering unit of the glomerulus had not yet been totally cleared.

So I think that's another message about why doses are important in terms of enzyme replacement therapy. I think the general blood vessel cells easily cleared, but the cells that are at the terminal parts of the circulation, such as those in the kidney glomerulus are not cleared. And so we have a chance to do something by increasing the enzyme. Now, parenthetically, in an entirely different conversation, it became very clear that if we're talking about cardiomyocytes or actually the podocytes themselves, the current dosing that we have available to us with the alpha-galactosidase alpha or the alpha-galactosidase beta, probably is not sufficient. We need something more, but that's kind of taking a glimpse into the future. This is what we had to work with in 2003. Now this slide is important to me for several reasons. One is that this represents the first patient treated with the commercial alpha-galactosidase beta in the US, and we're very proud of that in our UAB rare disease clinic.

Well, you'll see that over the subsequent time from 2003, that 2008, the patient did really well. That curve flattened out. That means the slope went from minus 13 units to only minus two units. So that was a substantial improvement. The plot itself stops in 2008. I haven't bothered adding all of the subsequent of the creatinines because the x-axis would stand out another three and a half feet. I didn't want to do that. But what I will tell you is that patient stabilized quite nicely. And finally, in 2022, two years ago, reached the point where he needed to have renal replacement therapy. And at that time he had a renal transplant and doing quite well. Parenthetically, his cardiac issues I think have been improved, but he was bothered by a persistent slow heart rate and has a pacemaker for that problem. So that's back to our notion that, well, I think what we have with our current enzyme replacement therapy is sufficient to deal with most of the patients with kidney issues, but may not ultimately be successful in terms of the cardiac involvement.

A final point about this patient that I'll share with you, for whatever reason, he never developed significant anti-drug antibody. And when asked what is the unmet need in Fabry disease, what do we need to focus on? My answer is the anti-drug antibody story. I think that has not been appreciated to the extent it needs to be, and I know you have personal experience with some of the infusion reactions that are related to the different kinds of anti-drug antibodies. A very interesting story, but something I'll chat about in a bit, which possibly might impact on that.

So here's my N of one, the story of enzyme replacement therapy as we know it. Now, remember, I'm a US practicing physician, so I used a lot of the approved alpha-galactosidase beta in our patients in US, but the other enzyme replacement therapy, alpha-galactosidase alpha, very popular, very useful in most patients in Europe and Canada, South America, Japan. I'm thinking patients, especially if they have milder disease, late onset variants, that sort of thing. I'm not sure there's an important distinction about

dose, but certainly in the patients who have kidney involvement, we felt very comfortable with aggressively treating the proteinuria and maximizing the enzyme replacement therapy.

Ozlem Goker-Alpan:

Obviously that original study for agalsidase beta included few different dosing regimens, and this was the dosing regimen that was deemed to be more safe, but obviously that the higher the dose, the better outcome, as far as I remember from that original study that when it was published. And Fabrazyme has been approved 18 years later, fully approved, as of March 2021 after its initial approval in 2003. Before we actually move into the second or new-generation enzyme replacement therapies, I think one of the more important points is obviously this patient predicted that the renal replacement therapy would occur if the rate of the renal decline had been obviously without treatment. But now what is the approach to a patient that comes with seemingly normal renal function, but we do now use more slopes of the renal decline rather than waiting the patient to have an abnormal creatinine? Is that right? Can you a little bit expand on that?

David Warnock:

Yes. The equation that we use to estimate the glomerular filtration rate, the so-called eGFR equation. There are a couple of them, and we can talk about which is your favorite flavor. It doesn't matter in my opinion, just so you use the same one and you have a valid serum creatinine measurement. But it includes the patient's age and the patient's gender in that calculation. So in a female especially, you can get back a serum creatinine value that says one in our standard units that we use here, and that's "normal." That's in the normal range. Well, that may be, but if it's a very small framed woman with not a lot of muscle mass, that could represent really a significant elevation in the creatinine. So that's why we use the estimated GFRs based on the serum creatinine rather than the serum creatinine by itself.

We've had an experience with daughters of our male Fabry patients. In Alabama, we've had a large number of the classic mutations which caused severe Fabry disease. We did not have as many of the late-onset mutations. In fact, we were not able to participate in the chaperone trials because we simply didn't have patients at that time that were appropriate for the chaperone therapy. But all right, daughters 15, 16, 18-year-old daughters of males who have Fabry disease. Well, they are, if you will, obligatory carriers. Obligate carriers, they have the mutation, you know that. They get one X from mom, one X from dad. Dad didn't give his Y chromosome to his daughter, he gave his X. And Fabry disease of course is X-linked. If you're a male, you have Fabry disease, the mutation is on your X chromosome, which you got from your mother, not from your father.

So we've had females at 15, 18 years of age come to us and say, "This is a terrible disease. I saw what it did to my uncle, I saw what it's done to my father. I want to know, would early treatment help me and when can we start?" Well, my answer to that is "Yes, of course. Great, let's do it. But I have a little problem with the insurance people at this point. I have to write a letter of medical necessity, and your serum creatinine level is normal. You don't have protein in your urine." So the response comes back, "Well, they're normal. Why do you want to treat these patients?" Well, the answer of course is, going back to our first slide that I showed you, they are in the cellular injury phase. We understand the progression of this disease, so we can demonstrate on renal biopsy that they have very significant involvement before there's permanent irreversible damage. So we've used that as a tool, an instrument to help us stage, help us understand when to start the therapy.

There's a similar story in the cardiac involvement of Fabry disease. We've had females, especially young females seen in our clinic who don't yet fulfill the criteria for left ventricular hypertrophy, which is the classic finding in Fabry disease. But we've proceeded to do the imaging to ask the question, could they

have fibrosis already without meeting the criteria for left ventricular hypertrophy? And we have found indeed that they can have this so-called late enhancement with the gadolinium scans before they have LVH. And so on the basis of that we've said, "Okay, let's start treatment. We have documented evidence that there's serious permanent damage that's already happened." We can't reverse that, but we hope to stabilize it. So the tissue, what we say in the renal world is tissue is the issue. The tissue tells a story, and we use those biopsies to help us understand the progress.

Ozlem Goker-Alpan:

So in genetic nomenclature, actually the Fabry disease taught us a big lesson. So we don't really call X-linked dominant or recessive anymore, but it's X-linked diseases. And we call the female patients with Fabry disease heterozygous patients with Fabry disease rather than carriers. Obviously in the past being named as carrier, like you experienced actually this franchise, this group of patients in a significant way. So let's talk about the new-generation enzyme replacement therapies and a little bit touch base with the unmet needs at the end, which you are very familiar of.

David Warnock:

Okay, so the new-generation, and again, we go to these meetings together, and I love these experiences because you come home with a hundred different ideas, and over the next week or two they kind of percolate. And the most important ones bubble up and they're at the top. So the interesting theme that has emerged now, the parlance seems to be the second-generation ERTs. Okay, what are second-generation ERTs? Well, it's very simple in my view. These are enzyme replacement approaches in which the enzyme has been stabilized. The normal enzyme in fact is a dimer. It has two identical units that are held together by electrostatic forces, and that stability is very important in the function of the enzyme. The active pocket is really between the two subunits, and if we can somehow stabilize that dimer, then the enzyme complex itself can have a longer dwell time, a longer half-life, if you will, and be more effective.

This is especially important when we are infusing ERTs, because they hang in a bottle of saline for three hours as we are infusing. And I think it's very clear that there's loss of enzyme activity in vitro, in the bag, before it gets into the patient. So stabilizing the enzyme is very important, and a couple of ways of doing that. There's efforts to manipulate the sequence itself to enhance the binding and stabilization under both the lysosomal conditions. That would be a pH, let's say, of 3.5, versus plasma pH of 7.4. But also there are a couple of examples now where the two monomers have been linked by a crosslinker of some sort, and that seems to stabilize it. The fundamental binding of the two monomers is driven by electrostatic forces, but if that dimer can now be stabilized by cross-linking, then we have, in the two examples I'm familiar with, a very prolonged half-life of the circulating infused enzyme in the circulation.

The traditional first-generation ERTs that we've talked about, alpha-galactosidase alpha and beta, those have plasma half-lives on the order of maybe 90 minutes, very short. The stabilized dimers, what I call the second-generation ERTs, have circulating half-lives in the patient, in vivo, in the plasma on the order of at least 45, 48, 80 hours. That's the time for half the enzyme to disappear. Tenfold, at least 10, maybe 20-fold increase over our first-generation. Well, is that important? My personal belief is yes, that's important. That means more enzyme is present longer for cellular uptake, for it to get to its targets to do what it's supposed to do. But that all needs to be proven of course by proper clinical trials and so forth and so on.

I will add, Ozlem, you and I had the pleasure of participating in a recent head-to-head comparison of the traditional first-generation alpha-galactosidase beta to one of the new-generations, the pegunigalsidase, or PRX-102, which was a head-to-head randomization, and the patients are treated with one mg per

kilogram every two weeks with either drug after having been on the traditional drug before they were randomized. The outcomes I think were encouraging. It looked as if the new second-generation was at least as effective as the traditional ERT, with the caveat of course that both drugs showed equivalent clearing of those deposits that we've talked about as being so important in the blood vessels. We can get into the weeds a little bit about the study design and what was actually shown. You and I have had these conversations more than once and have published them in a recent paper, so we're very pleased about that.

But this first example of the second-generation ERTs, as you mentioned, was approved actually... I'm sorry, it was approved last year, recent approved. The traditional ERT, the alpha-galactosidase beta, had its final full approval in 2021. This new second-generation was in 2023. There's an interesting... Now again, the comparison between the two was done at the same dose with the same dosing schedule. There's a second-generation ERT with which I am familiar, and which we both saw presented at the World Congress in San Diego two weeks ago. This has been stabilized with a small piece of the complement-fixing region of immunoglobulin, but it stabilizes the dimer. And in fact, this is being developed by a Korean company. It's not yet in human testing, human trials. So I'm really talking about preclinical data that has been published. I feel comfortable discussing that. I cannot guarantee that this is going to go forward and be finally approved in human, because the human studies have yet to be done, but I choose to be optimistic because this represents another opportunity for Fabry patients to have another option for their care.

But the interesting feature of this new agent is the company thinks that it can be developed for subcutaneous use once a month, which I think if this works for the patient would be such a boon. It's very clear that going to the clinic every two weeks for your enzyme infusion therapy, especially if you're talking about getting an adequate amount of enzyme, and hopefully you don't have an infusion reaction that is troublesome, but that's a chunk out of your life. And if you're a teenager in school or if you're trying to work and support a family or if you're a mom or a caregiver that has parents or children that are your daily responsibility, enzyme replacement therapy as we know it is a logistic challenge. So options to treat on a monthly basis I think would be really well received by the patient population.

We had an experience with that in an open label that is non-randomized experience with patients who were stable, doing well, and were then switched from the traditional every two week therapy to once a month therapy at twice the dose. And with a long-acting second-generation ERT, they appeared to do okay. But I have to tell you that as an unblinded experience, our patients were so enthusiastic, thought it was so wonderful that they didn't have to go to the clinic twice a month, they could do it once a month and even do that at home, that the interpretation of the results is colored by that enthusiasm, which is why we have to do a proper randomized study, a controlled study which will be needed before once a month therapy is approved for use in patients.

Ozlem Goker-Alpan:

So included in the label, is that right? It's not currently included in the labels, despite-

David Warnock:

It's not currently in the label. And because the FDA's point, as I understand it, was, well, you really didn't design the trial to convince us that this is the proper indication. And that's fine. Now, you and I both know that as a licensed physician, we can prescribe any licensed drug as we see fit, and I do think that there are groups of Fabry patients on ERT today that may not be well served by having it given once every four weeks rather than once every two weeks. I think we need more information before we can even come to any kind of firm conclusion about that. So I'm not willing to embrace, let's go with four

weeks therapy. I think we need a trial. We need to learn how to use these new second-generation drugs properly.

Ozlem Goker-Alpan:

Obviously it's the physician, the treating physician's sense clinic to decide on which patient will do better with different kind of treatment options? Is that right?

David Warnock:

That's absolutely correct, but I've been asked, as you have, many times about this, what's the driving, what's the decider? What decides are you going to switch from drug A to drug B? And I say, "Wait, wait, wait. Let's talk about the stakeholders. Who has skin in the game?" And I will tell you, if a patient comes to me and says, "I want to switch from drug A to drug B," I have to pay attention that because especially in our Fabry world, and I'm sure you see this in the other lysosomal diseases, you treat with, the patient and their families are very well-connected, very well-informed. They follow the treatment advances on a daily basis, so they have part of that decision making that is very important. I think the physician's experience and comfort zone is important, of course.

"Gosh, you're doing so well on the current drug. Why would you want to switch?" I mean, I can hear that question being asked. And I hate to even bring this up, but there are market forces at play and the insurance, the providers get involved in this now, and they do very interesting things. "Oh, of course, Doctor, this is approved. Certainly you can prescribe, but be aware that there are tier A drugs and tier B drugs, and the copayment for this drug is going to be $10 and the copayment for this is going to be $700." I mean, that kind of stuff happens. Don't ask me to defend it, please, but it's a fact of life. So this treatment decision, especially a switch, is a complicated conversation that-

Ozlem Goker-Alpan:

That is correct, and we are living as the day goes by. Before we talk briefly about the unmet needs, so you discussed actually that is the enzyme sitting in a bag and causing the little aggregation. Can we talk about a little bit, the new-generation ERTs on the infusion reactions, or how actually we have more than one treatment that is available with that idea? What is given in the chaperone that will actually... that is refold the protein in the bag, is that right, or when the patient gets infused?

David Warnock:

Right. There is a product actually for Pompe disease.

Ozlem Goker-Alpan:

Yes, exactly.

David Warnock:

Which is ERT combined with chaperone.

Ozlem Goker-Alpan:

Right. Yeah, exactly, yes.

David Warnock:

And the notion there of course is that you are stabilizing it in vitro. That's a very interesting concept.

Ozlem Goker-Alpan:

Exactly. Let's talk about this, the PEGylated products, about the aggregate formation, the infusion reactions, and the other immune response.

David Warnock:

Another feature of the second-generation drug that is currently approved, the pegunigalsidase alpha is its PEGylation. Now, in the product development and design, the company took huge care and responsibility, I would say, for designing the linking to make sure that it happens right. The first thing is that the dimerization I told you was driven by electrostatic forces. And at that point, they added a very short polyethylene glycol linker, three kilodaltons, and it was bifunctional. That meant both ends of it were active, both ends were looking for a binding partner, and this usually would be lysine, that if the two dimers are close together, I'm going to use these two fingers as the dimer representatives, see if I can do this. And here's my linker. That nanometric distance has to be just right for the linking. Once that's done, then they came back in with polyethylene glycol, three-kilodalton little monomers if you will, with only a single functional group.

And so these decorated the enzyme dimer on the outside. And I think, I can't remember, something like 12 of these PEG units were added. Well, PEGylation is very important, very well recognized as also increasing the stability of such a protein product in the infusion, in the circulation. There's another feature which is really interesting, and that is the PEGylation itself seems to impact on the antigen-presenting cells' recognition of epitopes. And so the possibility exists that if you don't have an antibody to your enzyme replacement therapy, and you're a classic patient that has no residual activity, that is you have no enzyme, the first time you're infused with the enzyme, the immune system is saying, "Whoa, wait a minute. That's something brand new. Never seen that before. I'm going to generate an antibody to it," which it does, as you well know, you're very experienced with some of the nasty ones.

At about the sixth or eighth treatment cycle, about three months into this, you start getting the phone call from the infusion nurses saying, "We got problems over here. The patient has fever, has chills, there's a rash, there's soreness at the injection site. They have terrible back pain." Well, this is the infusion reaction that we're familiar with. In a small sample, again, the phase one, phase two study, as I remember that trial, open label, I think it had 16 participants, two of them, both male, developed very significant antibodies against the enzyme replacement therapy, an anti-drug antibody. The titers kept going up, up, up, up, but then they stabilized and they came down. And in fact, they went almost to zero, to undetectable. So that's an example of something that we would call tolerization. And that's really exciting, because now that new antigen is being considered as self, and so the anti-drug antibody issue is not a problem.

In patients who already have anti-drug antibody and we switch to a different form of enzyme replacement therapy, there are all sorts of maneuvers at this point to try to minimize the anti-drug antibody reactions, but I am very pessimistic. Now, there is hope with the second-generations from two respects. One is if PEGylation is used, that can affect the immune modulation, if you will, the recognition of the antigen. The fact that the antigen itself, the infused ERT, is so stable and present for hours and hours and hours could possibly put the patient into an antigen antibody excess that could induce tolerance.

Ozlem Goker-Alpan:

That's correct.

David Warnock:

And I think that's what happens with enzyme-naive patients. There's also a risk that if the ratios aren't quite right, if you're in antigen antibody excess, you can actually develop immune complex disease. And that's a whole nother problem that I think we need to be at least aware of. I don't think it's very common, but it does happen. In fact, it's reported in the product insert for the new second-generation, so we have to be aware of it. But there's another possibility to... I think I would end up our conversation with the notion that can we modulate anti-drug antibodies if they're already formed, if they're already in place, what's new?

We've talked about using various immune modulation protocols and desensitization. You're very familiar with your IgA experiences and so forth. But there's a new agent that I'm working with, a company called Vera Therapeutics, and this is a very interesting decoy receptor. It binds to two of the most important cytokines that affect the control, both B-cell and plasma cell antigen recognition and antibody production. And the particular example that they're working on, IgA nephropathy, very interesting condition in which there's a glycosylation defect. So there's a stretch of the IgA1 that does not have proper glycosylation, and that becomes the site of a new epitope. So an antibody, usually IgG, is raised against the IgA1, and you now have an immune complex condition, IgA1 bound to IgG.

Now the physical characteristics of that complex are such that they deposit in the kidney in our friends the podocytes, and they get something called a mesangial proliferative glomerulonephritis. Well, it's a very common and severe form of kidney disease. Well, this new agent appears to reduce proteinuria in patients who've been resistant to other usual approaches to reducing proteinuria. It stabilizes the reduction, the fall in the kidney filtration rate so that it doesn't fall as fast or as far. But more importantly, and what really caught my attention, and when I was asked, "Are you interested in consulting with this about this?" I said, "Yes, on two conditions." I'll tell you in a minute. But the most exciting to me is that the new antibody, the IgA1 directed that has the glycosylation defect, that's the antigen, that is produced by plasma cells and is suppressed by this decoy receptor that actually binds to cytokines. So the actual bad antigen production is being suppressed.

And in that setting, to my mind, again, this is not approved and this is basic science stuff that we're talking about, but I think the prospect's there for all forms of immune complex diseases, and certainly worth discussing. And so I said to the company, "Jim, I'm really interested. I'm happy to help. I'm going to learn something from this for sure. But as my contribution, I want to tell you about IgA nephropathy." I'm sorry, "I want to tell your IgA nephropathy audience about Fabry disease, because we have an unmet need at this point with anti-drug antibodies. And if your approach, if your agent is such, proves in human trial to be as effective as you hope, I would think that we might start using this in some of our classic males who have serious anti-drug antibody problems."

Well, that's all speculation. It's based on published basic science work. This was presented at the American Society of Nephrology last year. But our patients, they always say, "Doc, what's the latest? When can I start taking the pill?" That's always been their favorite question, "When can I start the pill?" But as I always end my conversations, the patient conferences and discussions of individual patients to say, "Your disease is manageable, we can do a lot to help you, but research is on your side. New developments are coming all the time, and they're going to improve your outcome. And the chances that your children will have a normal life have gone up quite a lot since we got started on this voyage."

Ozlem Goker-Alpan:

Thank you, David, for both the historical perspective and this great scientific discussion, and I enjoyed tremendously. I'm sure our audience enjoyed as well. We would like to thank you again, and then we'll open the podium to the discussion and questions. Thank you.

David Warnock:

Great. Thank you for this opportunity. Appreciate it. Thank you.

Ozlem Goker-Alpan:

Thank you everyone who is still here for the discussion. There are actually one. There are a few questions, but as we are running off time, I will quickly answer. Dr. Warnock will be available via email to answer. So one question is how significant the females are affected in their renal involvement with Fabry disease? So females are not attenuated examples or the group with Fabry disease manifestations. They do have different disorder with different presentation, even though the kidney involvement, actually one of the organ involvement that could be more uniform. So when we look at the Fabry Registry data with the overall involvement, that includes classic and non-classic patient, that also involves females, about 41% of the patient population who are males, they would present with the chronic kidney disease stage three to five, and then this number is around half, about 20% of females that would have the similar CKD stages three to five in the Fabry Registry.

The data from Europe, that's when we actually think about the overall population at presentation, about one-third of the females will present with proteinuria. If you could recall, basically this is the environment of the podocytes, and there is already inflammation and cell fibrosis going on. And about, overall, 13% of the patients will have CKD stages three and above, and 4% of the patients who are female have end-stage renal disease. In our cohort, we follow quite a large cohort of patients, both males and females with Fabry disease, and almost male and female, loss of life is equal. So I need to attract attention for the nephrologists or for the physicians who are following female patients with Fabry disease to be vigilant in their follow-up and also the management and treatment. So I will close the podium, as we are running out of time. For the audience, I would encourage them to fill out the evaluation form, and we would like to thank the sponsor of this program, the companies that include Takeda, Sanofi, Chiesi, and Abacus. Thank you very much and have a great day.