[MUSIC PLAYING]

KATRINA HAN: Hi. My name is Katrina Han. I'm a clinical assistant professor in the Division of Endocrinology. Today, I'll be discussing lipodystrophy, which is a rare and often underrecognized syndrome that's associated with significant metabolic complications, so I thought it would be a helpful topic to review. I have no conflicts of interest or financial disclosures.

These are my objectives. I'm hoping that by the end of this talk, everyone will have a clearer picture of the clinical features and complications associated with each of the lipodystrophy syndromes; understand based on practice guidelines what is the appropriate approach to diagnosis of lipodystrophy; and finally, what are the recommendations for treatment of these patients? What's currently approved? And what are the potential therapies coming down the pipeline?

So first, I'd like to start with a case. This is a patient who was initially seen in the Pediatric Endocrinology Clinic, and later transitioned care to the Adult Endocrinology Clinic. So I'll go ahead and provide a brief summary of her initial clinical course.

She was born at a normal weight at term gestation without any perinatal complications. Mom was breastfeeding without any issues after her birth, but over the first few months, she had poor weight gain, dropping to the 25th percentile for weight by three months. So she was started on a 4-5 formula at that time.

However, she continued to have poor weight gain and drop to less than a 5th percentile weight by around six months of age. She was also having issues with recurrent bouts of diarrhea. And these episodes were typically accompanied by URI symptoms, or an ear infection. Of note, she did have a five-year-old brother who was going to daycare, so maybe bringing home whatever was going around at daycare at the time.

Otherwise, the rest of her development was intact. She was meeting milestones appropriately. Her pediatrician did make notice that on exam, she was very thin and quite muscular. At that time, he thought that perhaps her inadequate weight gain could be secondary to her recurrent illnesses and decreased oral intake during these episodes, but did feel that further evaluation was warranted, so she was referred to GI at the time.

They did a comprehensive assessment, which showed normal thyroid function, normal immunoglobulin levels, normal transaminase, GGT, and alk phos, but she was noted to have an elevated triglyceride level, at 224, with a mildly low HDL at 31 and a normal LDL. And given the presence of hypertriglyceridemia at such a young age and her appearance on exam being very thin and muscular, the GI provider was concerned for lipodystrophy.

They then checked complement C3 and C4 levels. And some children with lypodystrophy can have hypocomplementemia. For her, these ended up being normal. She also had a normal fasting glucose and normal glucose tolerance tests, but her fasting insulin level was noted to be elevated. With time and continued monitoring, her diarrhea resolved without any intervention and her weight did improve. So by the age of two, she was falling between the 50th and 75th percentile for her weight.

Her exam at the time was notable for hypertrichosis and acanthosis nigricans. Her A1C was 4.7%. After that, she seemed to do OK for several years without any major changes, until she turned 10 years old. At that time, her A1C increased to 5.5%, then 6.1%.

She was also noted to have hepatomegaly and was referred back to GI for this. A liver ultrasound demonstrated fatty change and enlargement. She had normal serum transaminases at that time. On exam, she had marked absence of subcutaneous fat, with thickening of her skin at the neck and axilla. Diet and lifestyle modification were recommended at that time.

At age 13, diabetes was confirmed on an oral glucose tolerance test. Her triglycerides had increased to 643, with an LDL of 93. She was started on metformin, as well as fish oil for this, and she started having periods which were irregular and was seen by gynecology, who suspected PCOS. She had a pelvic ultrasound, which had large ovaries and also a cyst. And she was started on oral progesterone for this.

Unfortunately, her diabetes was uncontrolled, despite escalating doses of insulin. She was up to 44 units of Lantus at one point. And there were some concerns about intermittent medication noncompliance, but it seemed even when she was taking her medications, there was still difficulty keeping her blood sugars controlled. So we will circle back to her case to go over what was done at that time closer to the end of this talk.

So just to get everyone thinking a little bit more, I wanted to pose a few questions. So based on this case presentation, what type of lipodystrophy does this patient have? What additional testing is indicated for this patient? And what is the recommended approach to treatment and long-term monitoring for this patient, based on the clinical practice guidelines? So these are some of the questions that I will be providing the answers for as I go through the rest of my presentation.

But first, since we are talking about lipodystrophy, it is important to go back to some basics. So starting with adipose tissue, it was initially thought to be a passive reservoir for energy storage, but we now know that it is actually a highly active endocrine organ that expresses and secretes a variety of bioactive peptides, known as adipokines, which have both local and systemic effects.

As early as 1878, it was identified as a major site for metabolism of sex steroids. And by 1994, we learned that it was the source of leptin. In addition to these efferent signals, adipose tissue also expresses a number of receptors which allow it to respond also to afferent signals from traditional hormone systems, as well as the central nervous system. Therefore, it plays an integral role in energy metabolism, neuroendocrine function, as well as immune function.

Leptin, as I mentioned, was first discovered in 1994. And this was done by Dr. Jeffrey Friedman and his research group at the Rockefeller University, who you see here on the right side. Prior to this discovery, parabiosis studies in mice were carried out, which really provided the data to show that there were some sort of circulating factor that could communicate information about the sufficiency of energy stores between the periphery and the central nervous system.

These initial studies were conducted by Dr. Douglas Coleman, who you see on the left here, at the Jackson Laboratory in the 1960s. So both Dr. Coleman and Dr. Friedman share the 2010 Albert Lasker Basic Medical Research Award for their contributions towards the discovery of leptin.

So what is parabiosis? It is the surgical union of two organisms-- in this case, mice-- allowing them to share blood circulation. The specific parabiosis studies that helped to lay the groundwork for the discovery of leptin used mice carrying recessive mutations in the diabetes or DB gene, which resulted in a phenotype of obesity, hyperphasia, polydipsia, and polyuria. And then also mice with recessive mutations in the obesity, or ob gene, which resulted in a phenotype of obesity and hyperphasia.

In these studies, Coleman paired the db/db mice with wild type mice. And what ended up happening was, for the wild type mice, instead of overeating like the db/db mice did, they actually stopped eating and died from starvation. So this confirmed Coleman's hypothesis, that the db/db mice must release some sort of factor that inhibits the drive to eat, which is what caused the wild type mice to starve, but that the db/db mice cannot respond to the signal themselves. So that's why they continued overeating.

When you pair the db/db mice with the ob/ob mice, the ob/ob mice actually stopped eating and starved to death, while the db/db mice, again, remained obese. In contrast, attaching the ob/ob mice to the wild type mice actually did nothing to the wild type mice, but it did cause the ob/ob mice to limit their food consumption and actually gain less weight. So putting these findings together, Coleman concluded that the ob/ob mice failed to produce this circulating factor that inhibits eating, while the db/db mice overproduce this factor, but lack the receptor to be able to respond to it. Many years later, it was discovered that leptin was this factor, so leptin was encoded by the ob gene, while the leptin receptor was encoded by the db gene.

So we now know that leptin is a peptide hormone secreted by adipose cells in proportion to overall body fat mass. So in a well-fed state, there is increased fat, which leads to increased leptin secretion. It enters the central nervous system and initiates a JAK-STAT secondary messenger pathway in the hypothalamus, which leads to decreased food intake and increase energy expenditure to maintain homeostasis in response to high fat levels. Leptin decreases glucose stimulated insulin secretion from the pancreas; decreases gluconeogenesis in the liver; and in the muscle, it increases glucose uptake and increases fatty acid oxidation, which leads to decreased lipid accumulation.

Leptin also plays a role in regulation of the menstrual cycle. And it also has several other important roles that are not pictured here, including regulation of immune function, hematopoiesis, angiogenesis, as well as bone development. So having a knowledge of this background information will allow us to better understand lipodystrophy, and why we see the metabolic complications often associated with it.

So what is lipodystrophy? Well, it is a heterogeneous group of rare, congenital, or acquired disorders characterized by either complete or partial loss of subcutaneous adipose tissue. They can result from either failure of adipocyte development or the immune-mediated premature destruction of adipocytes.

The lack of adipose tissue storage capacity is associated with lipid accumulation in ectopic sites such as the liver, muscle, pancreas, and kidneys, leading to the many metabolic abnormalities that we see in patients with lipodystrophy. And not only is there a loss of storage capacity for excess energy, but there's also a reduction in the levels of adipokines, notably leptin, which often leads to multiple metabolic abnormalities as well.

The extent of fat loss typically collates with the severity of these metabolic abnormalities, which goes to show the importance of adipose tissue as an active endocrine organ. But not only is the total amount of adipose tissue present an important factor, we also see that the appropriate distribution of fat deposits plays a significant role in metabolic function as well.

So in terms of the clinical characteristics and comorbidities that we often see in lipodystrophy, patients often have increased appetite and are hyperphasic. They get muscle hypertrophy and often have prominent veins, since they lack subcutaneous adipose tissue. They develop acanthosis nigricans in the setting of insulin resistance. Women can also have polycystic ovaries and ovarian dysfunction with hyperandrogenism.

Patients are unable to store extra energy in subcutaneous fat tissue, so they get fat accumulation in other organs, including the liver, leading to fatty liver and hepatic steatosis. They get hypertriglyceridemia, which can lead to pancreatitis or eruptive xanthomas, as well as other associated comorbidities, including cardiovascular disease. There have also been reports of nephropathies characterized by proteinuria and hyperfiltration in patients with generalized lipodystrophies.

This was a study published in JCEM, which examined serum adiponectin and leptin levels in patients with four different types of lipodystrophy. And I will be discussing the differences between these different types in the next several slides. This study enrolled a total of 93 patients-- 18 with congenital generalized lipodystrophy, 11 with acquired generalized lipodystrophy, 46 with familial partial lipodystrophy, and 18 with acquired partial lipodystrophy. And what they found was that serum adiponectin and leptin levels were lower in patients with congenital or acquired generalized lypodystrophy, which are the two types characterized by near-complete absence of subcutaneous adipose tissue. You can also see that the fasting insulin levels tended to be higher, with greater rates of diabetes in these two types as well.

So as I mentioned, there are different categories of lipodystrophy syndromes and also subtypes within each of these categories, all with their own unique clinical findings. So for these four main buckets, they are categorized based on etiology-- so whether it's genetic or acquired-- and also based on the distribution of loss of adipose tissue. So whether it's generalized or partial loss of adipose tissue.

So this gives us the four main categories of lipodystrophy syndromes, as you see listed here. And I'll go over each of these and some of the unique findings for each one. So starting with congenital generalized lipodystrophy, also called Berardinelli-Seip syndrome, named after the physicians who described the first cases. It is an autosomal recessive disorder, with a prevalence of about 1 in 10 million people, so very rare.

In these patients, you start to see the absence of subcutaneous fat very early on, typically at birth or within the first two years of life. Adipose tissue is almost completely absent from most areas, including the abdomen, thorax, arms, and legs. But there's often normal amount of adipose tissue around the orbits, the mouth and tongue, the palms and soles. So these areas are often spared.

They often have huge appetites in childhood, with accelerated linear growth, increased metabolic rate, as well as advanced bone age, although their final height is usually normal. They have very prominent muscles, develop acanthosis nigricans, hepatomegaly, and also tend to have acromegalic features, with enlarged mandibles, hands, and feet, which is the sequelae of elevated circulating insulin levels leading to soft tissue and bony overgrowth.

Multiple genetic causes have been identified, with the most common ones being mutations of the AGPAT2 gene, which encodes an enzyme involved in the biosynthesis of triglycerides and glycerol phospholipids, and BSCL2 gene, which encodes a protein called seipin, which appears to be critical for normal adipogenesis in vitro. These are responsible for 95% of reported cases. Metabolic complications are frequent and may be severe.

The next major subtype is familial partial lipodystrophy, also called Kobberling-Dunnigan syndrome. So this is a group of usually autosomal dominant disorders occurring in about 1 in one million people. Typically, these patients have loss of subcutaneous fat, mostly from the extremities, the buttocks, and the hips. Fat distribution is usually normal in early childhood, with loss of fat starting to occur around puberty.

There's often some regional excess fat accumulation. So for example, in this patient, you can see she has some accumulation of subcutaneous fat in the face, as well as in the chin. Muscular hypertrophy, again, is common. And there have been multiple genes identified to be associated with the different subtypes of familial partial lipodystrophy, as you see listed here. Some of these gene mutations lead to, for example, premature death of adipocytes, abnormal regulation of adipogenesis, lipolysis, and lipid storage, as well as reduce adipocyte differentiation. Metabolic complications are common in adulthood, including increased risk of coronary heart disease.

So now moving on to the acquired type-- so starting with acquired generalized lipodystrophy, or also called Lawrence syndrome. This occurs more frequently in females and in males, with a ratio of about 3:1. Only about 100 cases have been reported. And they have characteristics which are similar to those of patients with congenital generalized lipodystrophy. However, the big differentiating factor here is the timing of onset.

So this occurs in a previously healthy child or adult. And typically, changes occur over the course of days to weeks. It usually happens before adolescence, but really it can develop at any point in life, with progressive loss of fat affecting the whole body, often including the palms and soles. So that's how is different from congenital generalized lipodystrophy, where you often see sparing of the palms and soles. Some fat accumulation can appear in the face, neck, or axilla.

It is not always clear what the trigger or cause is, but based on some reports, it might be triggered by an infection. In some cases, the development of antibodies against adipocyte membrane antigens have been detected, and the syndrome is often associated with other autoimmune diseases. Again, metabolic complications are frequent and may be severe.

So acquired partial lipodystrophy, also called Barraquer-Simons syndrome, is also more frequent in females and males, with a 4:1 ratio. There have been about 250 cases reported. And it usually appears starting in childhood or adolescence, often after a febrile illness, then over the course of months to years, there's a gradual loss of fat starting from the head and then moving downwards, progressively affecting the face, then the neck, the shoulders, arms, and trunk. Fat accumulation can occur in the hips, buttocks, and legs.

In terms of pathogenesis, it seems to be related to accelerated complement activation and the presence of C3 nephritic factor, which is an immunoglobulin thought to cause lysis of adipose tissue that expresses adipsin. So it's thought that this specific pattern of fat loss is seen due to the heterogeneity of adipsin expression at different sites of adipose tissue. It's also associated with autoimmune diseases, especially membranoproliferative glomerulonephritis, which occurs in about 20% of these patients. Some have acanthosis nigricans or clinical manifestations of ovarian hyperandrogenism, but the prevalence of diabetes is much lower for these patients compared to the other forms of lipodystrophy.

So metabolic complications are actually uncommon in these cases. I did want to mention one more subtype, and that's HIV-associated lipodystrophy syndrome. It's thought to be related to antiretroviral therapy, and is distinct from HIV-related wasting, felt to be caused by either the HIV infection itself or opportunistic infections and cancers which lead to the waste of not only the adipose tissue, but also other tissues, such as muscle.

This is actually currently the most prevalent form of lipodystrophy. It's estimated to occur in about 13% to 34% of patients on HART, typically when they've been on treatment for at least two or more years. It worsens with ongoing therapy and does not typically reverse with this continuation of treatment. It is more prevalent in males and is estimated to affect about 100,000 individuals in the United States.

You typically see a pattern of fat loss from the face, arms, the legs, and buttocks, which can occur along with fat accumulation in the abdomen and sometimes the dorsal cervical area. Risk factors for this include older age, greater severity of HIV infection with increased viral load, low CD4 count, and co-infection with hepatitis C. At this time, the mechanisms for how the antiretroviral therapies contribute to lipodystrophy are not completely understood. It is thought that the NRTIs may contribute to mitochondrial toxicity, characterized by abnormal changes in mitochondrial proliferation, morphology, and DNA content.

Protease inhibitors have also been shown to disrupt adipocyte differentiation via downregulation of several adipogenic transcription factors, like PPAR gamma and CEBP alpha. Compared to protease inhibitors and NRTIs, there's less direct evidence that the NNRTIs cause lipodystrophy or other metabolic changes. In these patients, we do see an increased risk for insulin resistance, diabetes, dyslipidemia, as well as cardiovascular disease.

So how do we diagnose lipodystrophy? Well, it's a clinical diagnosis based on history, physical exam, and metabolic status. There's not any defined leptin level that establishes or rules out the diagnosis. And that's because serum leptin assays are not standardized. Some patients with lipodystrophy, especially partial forms, have leptin levels similar to those of the general population.

So AACE has provided the following guidelines for when to suspect lipodystrophy. They describe the core clinical characteristics as the loss or absence of subcutaneous body fat in a partial or generalized fashion, with supportive findings that include the presence of diabetes with severe insulin resistance, ketosis-resistant diabetes, acanthosis nigricans, or signs of hyperandrogenism in females. Also, hypertriglyceridemia that is severe and not really responsive to therapies, or history of pancreatitis as a complication of hypertriglyceridemia. They also include evidence of hepatic steatosis based on hepatic magli on exam, or on imaging with or without elevated transaminases. Other family members with similar physical appearance with fat loss, the presence of prominent muscles and enlarged veins, hyperphagia, secondary hypogonadism in a male, and primary or secondary amenorrhea in a female.

So this is the diagnostic approach provided by the Endocrine Society guidelines. So when you do suspect lipodystrophy, it's important to get a good history exam, as well as labs to evaluate for metabolic abnormalities. And I did not mention this yet, but there are some progeroid syndromes that are associated with genetic lipodystrophy. So the presence of progeria features would make you lean towards a genetic subtype, whereas the presence of an autoimmune disease would increase the suspicion for an acquired lipodystrophy.

If there's a family history of lipodystrophy, that would push you again towards a genetic type. If there's not a family history of this, then it's helpful to consider the timing of onset. So with onset at birth or infancy, that would again push you towards a genetic type. If onset is in late childhood or after, then it's helpful to consider the distribution of fat loss. So if loss of fat is primarily seen in the extremities, that would push you towards genetic.

If you recall, this is the pattern typical of familial partial lipodystrophy. And if the fat loss is from the upper body only or generalized, this would be more consistent with an acquired lipodystrophy. So we're thinking it might be genetic. Genotyping can be helpful to confirm the diagnosis, but of note, a negative test does not rule out lipodystrophy, since there is evidence that additional loci for genetic dystrophies exist that we may not know enough about yet.

When suspecting an acquired lipodystrophy, getting additional labs may be helpful to support this diagnosis. And some of the findings that would be supportive include a low serum C3; the presence of C3 nephrotic factor; positive proteinuria; or biopsy-proven membranoproliferative glomerulonephritis, which, again, is associated with acquired partial lipodystrophy.

So following diagnosis, the recommended management of these patients from the Endocrine Society guidelines include regular screening for metabolic abnormalities and complications. So this includes annual screening for diabetes. Lipid panel should be checked annually and as clinically indicated. For example, with abdominal pain, that might be concerning for pancreatitis-- or with the presence of erupted xanthomas. Liver transaminases should also be checked annually, with a liver ultrasound done at the time of diagnosis and then as clinically indicated.

Annual uterine protein is also recommended. And in terms of screening for reproductive dysfunction during childhood, pubertal staging should be performed annually, as children with generalized lipodystrophy can have early adrenarche, true precocious puberty, or central hypogonadism. For cardiovascular health, blood pressure should also be checked annually. And in cases of congenital generalized lipodystrophy, the guidelines do also recommend annual ACG and TTE.

So management involves treatment of the metabolic disturbances. So treating diabetes and hypertriglyceridemia using the same methods you would in patients without lipodystrophy-- so with diet; lifestyle modification; medications such as metformin, statins, and/or fibrates. Insulin or other oral agents may also be used.

Now, if the metabolic issues are still persistent despite the use of more traditional therapies, then another option is the use metreleptin, or Myalept. So this is a recombinant human leptin analog. It was approved by the US FDA in 2014 for use in patients with congenital or acquired generalized lipodystrophy. Safety and effectiveness have not yet been established for treatment in partial lipodystrophy, so it's not approved for those patients at this time. And actually, metreleptin was initially studied as a potential treatment for weight loss. However, the data did not show significant improvements or benefit when used for treating patients with obesity.

Metreleptin is a once-daily injection that comes as a powder and requires reconstitution with sterile water before injection into the upper arm, thigh, or abdomen. It must be stored in the refrigerator, and once reconstituted, should be used within three days. It's currently only available through this risk evaluation and mitigation strategy program. Physicians must be enrolled and certified in this program before they can prescribe metreleptin.

The most common side effects seen in clinical trials have included headaches; hypoglycemia, especially when patients are on insulin; decreased weight; and abdominal pain. And there are a couple of boxed warnings that I will be talking about. But first, I want to show the outcome's data, which have demonstrated the efficacy of metreleptin.

So this was a study published in *The New England Journal of Medicine* in 2002. This was a prospective open label study at the NIH and UT Southwestern. They enrolled nine patients. All were female and, as per inclusion criteria, had serum leptin level less than 4. Patients were treated with metreleptin for four months and assessed for improvements in insulin resistance, diabetes, and hypertriglyceridemia.

So this table summarizes the baseline clinical characteristics of the nine patients treated in the study. The patients ranged from 15 to 42 years of age. Five out of the nine had congenital generalized lipodystrophy, three had acquired generalized lipodystrophy, and one had familial partial lipodystrophy. Over the course of the study, doses of hypoglycemic drugs were tapered or discontinued as needed.

Eight of the nine patients had diabetes. And all of these eight patients were receiving medications for their diabetes before the study began. Four of the nine were on lipid-lowering therapy. Mean serum leptin level was 1.3 at baseline and increased 11.1 at the end of four months of treatment. Glycemic control improved despite the fact that antidiabetic therapy was only decreased or discontinued during the four months of treatment.

You can see there was a significant de-escalation of therapy with patients being able to stop their very high doses of insulin. Here, you can see the individual metabolic values from before and after each month of treatment through the end of four months. They do mention compliance issues with patient two between months three and four. And patient nine did not have diabetes. But otherwise, you can see that each patient had significant improvement in their fasting plasma glucose, A1C, as well as fasting plasma triglycerides with treatment.

I wanted to go over another study that was conducted by Dr. Rebecca Brown and her research group at the NIH, demonstrating the effectiveness of metreleptin in the treatment of congenital generalized lipodystrophy. And this was another open label study that ran from 2000 to 2014. They included data from a pilot study and its longterm extension, which were integrated into one final analysis. They enrolled a total of 66 patients. 77% were female, with ages ranging from six months to 68 years old. 68% had congenital generalized lipodystrophy, whereas 32% had acquired generalized lipodystrophy.

The pilot study required subjects to have leptin levels less than 4 for females and less than 3 for males, whereas the longterm study required leptin levels less than 12 for females and less than 8 for males. Subjects had to have at least one metabolic abnormality, including the presence of diabetes as per ADA criteria, fasting insulin greater than 30, a fasting triglyceride greater than 200. The primary efficacy endpoints included change in hemoglobin A1C and fasting triglycerides at month 12. They did also assess for change in fasting plasma glucose, changes in insulin, and other antidiuretic or lipid-lowering medications, as well as change in liver size based on MRI imaging as a marker of fatty liver disease.

And what they found was that treatment with metreleptin led to significant improvements in glycemic control and hypertriglyceridemia over 12 months, with A1C reduction from a mean baseline of 8.6% down to 6.4%, and fasting triglycerides concentrations from 1,300 from baseline down to 398 after 12 months, or about a 32% reduction. And those are numbers I converted to milligrams per deciliter, which is different from the millimoles per liter you see in the bar graph. Fasting plasma glucose decreased from 184 to 126.

You can see there are two sets of bars for these three different graphs. And the first set of bars represents the full analysis set, while the second set of bars on the right side of each one represent the controlled concomitant medication full analysis set. So these include patients whose antidiuretic or lipid-lowering medications were either unchanged or only decreased from baseline-- so to try and isolate for the effects of metreleptin alone. And you do see that the improvements are still present, even after accounting for the medication changes.

There was also a significant decrease in liver volume by about 34%. And this was estimated based on MRIs, suggesting an improvement in hepatic steatosis, since that does correlate with liver volume. In the study, there were significant changes in these serum markers at all time points, including at six months, 12, 24, 36, and 48 months, without any loss of efficacy over time. And they reported seeing improvement as early as within the first week of treatment.

They also assessed safety endpoints, including duration and dose of metreleptin exposure, incidents of treatment-emergent adverse events, and serious adverse events. So this includes the summary of adverse events in the safety analysis set. The most common adverse events were consistent with the expected effects of metreleptin, including decrease in weight, hypoglycemia, and decreased appetite. Mean change in weight was a loss of 2.2 kilograms from baseline to month 12. Patients who experienced hypoglycemic events were on insulin therapy with or without oral agents.

There were three patients with reported serious adverse events. One had severe respiratory distress associated with chest pain and dyspepsia, and another with a history of asthma who had severe chest discomfort, flushing, and dyspnea with related panic attack. In both of these cases, metreleptin was able to be resumed without any recurrence of any adverse reaction. A third patient experienced cardiac arrest as a likely consequence of pancreatitis and septic shock, and this was deemed unrelated to study treatment.

So overall, in the 14-year study duration, treatment with metreleptin was generally well-tolerated, with most treatment-related adverse events being only mild to moderate in severity. So one thing to note is that metreleptin-- the US label has two boxed warnings. The first is about neutralizing antibodies. We know that in general, the development of antibodies against therapeutic proteins occurs, but may not always have any clinical relevance.

However, sometimes they can lead to loss of efficacy by affecting the pharmacokinetics and neutralizing drug activity, or it could also neutralize the endogenous counterpart of what the drug is targeting. So this is something that actually has been seen with metreleptin. There have also been cases of severe infections that occurred with the development of these anti-metreleptin antibodies.

So looking at the patients from the same NIH open label study mentioned before, there were 43 patients that they obtained anti-metreleptin antibody data on. Of these patients, 36-- or about 84%-- developed antibodies during treatment with metreleptin, with titres ranging from 1 to 5 to 1 to 78. The duration of treatment at the time of antibody assessment ranged from 4 to 138 months. Most patients remained antibody positive throughout the treatment course, but titres generally decreased over time despite continued metreleptin exposure.

Of the 36 patients that developed these antibodies, only four patients developed signs suggesting a loss of effectiveness of metreleptin. And this was based on worsened glycemic control and increased triglycerides. However, given the small number of reports, the clinical implications associated with these neutralizing antibodies really are not well-characterized. But it is recommended that patients be tested for these anti-metreleptin antibodies if they do start showing signs that the medication is no longer effective.

So the other box warning is regarding T-cell lymphoma. There have been three cases of T-cell lymphoma reported in the MYALEPT REMS program. All three patients had acquired generalized lipodystrophy. Two of these patients were diagnosed with peripheral T-cell lymphoma while receiving metreleptin. But of note, both had immunodeficiency and hematologic abnormalities, including severe bone marrow abnormalities, even before starting metreleptin.

There was one separate case of anaplastic large cell lymphoma, which was reported in a patient receiving metreleptin who actually did not have any hematologic abnormalities prior to treatment. Because leptin receptors are expressed on T cells, there is concern that metreleptin could play a role in lymphoma development. However, lymphomas in acquired generalized lipodystrophy have occurred both with and without metreleptin treatment, so metreleptin may not be playing a primary role in this.

However, its theoretical role on tumor growth cannot be ruled out. So it is something to consider and to discuss with patients, especially if they have a history of any hematologic abnormalities. But still, for most patients, the proven benefits of metreleptin will likely outweigh the theoretical risks of lymphoma.

So the same research group also published data on the effects of metreleptin in patients with partial lipodystrophy. So this was similarly a prospective, non-randomized open label clinical trial conducted at the NIH. They enrolled 41 patients. 98% were female. Ages ranged from 10 to 64 years old. 85% had familial partial lipodystrophy and 15% had acquired partial lipodystrophy.

Inclusion criteria were the same as in the previous study, with leptin less than 12 in females and less than 8 in males, with at least one metabolic abnormality. They measured the same outcomes with change from baseline to the end of month 12 in A1C, fasting triglycerides, fasting plasma glucose, and liver volume. The results showed that treatment with metreleptin did lead to significant reductions in A1C from amino 7.9 at baseline to 7.4 at the end of 12 months. For fasting triglycerides, there was a reduction from mean baseline of 1106 milligrams per deciliter to 477, which was about a 20% reduction.

Fasting plasma glucose decreased from 158 to 135. And again, I'm saying numbers that I converted to milligrams per deciliter, since they use millimoles per liter in their graphs. Liver volume decreased by 13.4%. So there was improvement with treatment, although not to the degree we saw in patients with generalized lipodystrophy.

They also did a subgroup analysis of patients who had baseline A1C of 6.5 or greater and/or fasting triglycerides of 500 or greater-- so those with more severe metabolic disease. And this included 31 out of the 41 patients. The results showed greater improvement, including decrease in A1C from 8.7 at baseline to 7.9, a decrease in mean triglycerides by 37.4% from a mean of 1389 to 530.

And fasting plasma glucose decreased by about 13.2%, from 180 to 146. Liver volume decreased by 12.4%. So overall, this study showed that metreleptin appears to be beneficial in the treatment of partial lipodystrophy as well, and likely more beneficial for a subset of patients who have more severe metabolic disease.

I wanted to talk about one other open label study that was also conducted at the NIH and published in JCEM a few years ago. As I mentioned, one of the complications that has been seen in lipodystrophy includes proteinuric nephropathies. A variety of pathologic changes have been reported in lipodystrophy, including glomerular hypertrophy, mesangial expansion, podocyte injury, diabetic nephropathy, FSGS, and membranoproliferative glomerulonephritis. The mechanisms behind these changes seem to be complex and not yet fully understood.

So they enrolled 115 patients. 83% were female with non-HIV-associated lipodystrophy. 73 patients had generalized lipodystrophy and 42 had partial lipodystrophy. They used similar eligibility criteria as the previous studies, including the same cutoffs for leptin levels and at least one metabolic abnormality. Outcome measures included 24-hour urinary albumin and protein excretion rates, eGFR, and creatinine clearance, which were all measured at baseline, and during up to 24 months of metreleptin treatment. Patients with increases in medications, including ACEs and ARBs, affecting outcome measures were excluded.

And what they found was that at baseline, patients with generalized lipodystrophy had significantly greater albuminuria, proteinuria, eGFR, and creatinine clearance, compared with patients with partial lipodystrophy, as you can see from the graphs here. So the graphs in the left column show the albumin excretion rate, protein excretion rate, eGFR, and creatinine clearance for patients with generalized lipodystrophy, while the graphs in the right column show those measurements for patients with partial lipodystrophy. You can see that, with up to 24 months of metreleptin treatment, there were significant reductions in albuminuria and proteinuria in patients with generalized lipodystrophy, but not in patients with partial lipodystrophy.

No changes in eGFR and creatinine clearance were observed in patients with generalized or partial lipodystrophy during metreleptin treatment. So to put these findings into perspective, a recent meta analysis had demonstrated that in patients with diabetic nephropathy, a sustained 30% reduction in albuminuria predicted lower progression to end stage kidney disease. And we do see that for patients with generalized lipodystrophy who were treated with metreleptin-- reach and even surpass this threshold in reduction.

But it's not yet clear whether this improvement will directly translate to the reduced risk of end stage renal disease in these patients. As I mentioned, there are various renal abnormalities, not just diabetic nephropathies, seen in patients with lipodystrophy. And furthermore, it wasn't clear whether the reduction in proteinuria with metreleptin was due to direct effects of leptin on the kidneys or due to an overall improved metabolic state with metreleptin treatment.

So what about the effects of metreleptin on survival? So this was a study published in JCEM a couple of years ago that examined the effects of metreleptin on survival of patients with non-HIV-associated lipodystrophy, including both generalized and partial lipodystrophy. Retrospective data were collected on 105 patients, with generalized lipodystrophy and partial lipodystrophy who received metreleptin as part of the studies I previously discussed from the open label studies that took place at the NIH between 2000 and 2014.

Separately, data from patients who never received metreleptin, based on medical records of 230 patients obtained from multiple centers, including at the NIH, University of Michigan, University of Sao Paulo, Brazil, as well as one other university in Brazil and one in Turkey. They matched patients based on age, sex, whether they had generalized versus partial lipodystrophy, the number of organs among the heart, liver, and kidneys with an observed abnormality, and elevated hemoglobin A1C levels of 6.5 or above.

They did mention that, even after matching, some metabolic abnormalities were more prevalent in the metreleptin-treated cohort, due to an inherent bias towards treating more severely affected patients. In the metreleptin-treated cohort, there were 11 deaths among patients with generalized lipodystrophy and one death among patients with partial lipodystrophy. In the metreleptin-naive cohort, there were nine deaths among patients with generalized lipodystrophy and three deaths among patients with partial lipodystrophy. The most frequently reported causes of death as recorded were heart, liver, or kidney disease, or infections. And in general, these are usually the most common causes of death in patients with lipodystrophy.

Kaplan-Meier analysis did not reveal a statistically significant difference in time to mortality between metreleptin-treated patients when compared with matched metreleptin-naive patients. However, results of a Cox proportional hazards model show that, after adjusting for other covariates, metreleptin treatment was associated with a 65% reduction in mortality risk. So while the study did have limitations, including the low number of mortality events, the findings do suggest that metreleptin may result in reduced mortality risk for patients with lipodystrophy.

Currently, there are other ongoing trials for potential treatment options other than metreleptin for lipodystrophy. And one of these studies is actually being conducted here at UPMC by Dr. Erin Kershaw, who's conducting a randomized double-blind placebo-controlled study examining the effects of a leptin receptor agonist antibody in the treatment of familial partial lipodystrophy. Another agent that has been under investigation is an antisense oligonucleotide inhibitor of apoC-III, which I'll talk a bit more about, and also an antisense inhibitor of ANGPTL3, which I'll also be talking a little bit more now.

So volanesorsen, or Waylivra, is an antisense oligonucleotide inhibitor of apolipoprotein C-III mRNA. So we know that apoC-III inhibits lipoprotein lipase. And patients who have genetically low levels of apoC-III have lower triglyceride levels and reduced rates of cardiovascular disease. So this medication was actually approved in Europe in May of 2019 for treatment of adults with familial chylomicronemia syndrome, which we know is caused by mutations in lipoprotein lipase, resulting in severe hypertriglyceridemia.

And the NIH recently conducted a study to assess the effects of this medication in patients with partial lipodystrophy. So this was a phase II randomized double-blind placebo-controlled study that took place between 2015 to 2019. They presented some of their findings as an abstract submitted to the Endocrine Society in May a couple of years ago. They enrolled five adults with partial lipodystrophy and triglycerides of at least 500 or greater, or triglycerides of 200 or greater with an A1C greater than 7%. They received either treatment with volanesorsen or placebo for 16 weeks, followed by a 1-year open label extension.

After 16 weeks of treatment with the active drug, they saw a decrease in mean apoC-III levels from 380 to 75. Triglyceride levels decreased from 503 to 115 and there was an increase in lipoprotein lipase activity. There was no significant change in A1C, however there was a decrease in HOMA-IR, from 26 to 13, and an increase in peripheral insulin sensitivity, as well as hepatic insulin sensitivity, based on the results of clamp studies. Adverse events that they noted included injection site reactions and decreased platelets, though they didn't go into further details than this. So this seems to be one potential treatment option that we could see become available sometime in the future.

Another therapy that has been undergoing investigation is vupanorsen, which is an N-acetyl galactosamine-conjugated antisense oligonucleotide targeted to the liver that selectively inhibits into ANGPTL3 protein synthesis. We know that ANGPTL3 is an inhibitor of lipoprotein lipase. And previous studies have shown loss of function mutations in ANGPTL3 are associated with reduced risk of coronary artery disease and reduced risk of insulin resistance in diabetes. So this drug was designed to lower ANGPTL3 levels.

In the phase I and II clinical studies, volunteers with elevated triglyceride levels were treated with this drug. And the results were published in *The New England Journal of Medicine* in 2017, which showed that the treatment resulted in dose-dependent reductions in triglycerides, LDL, apoB and apoC-III protein. Unfortunately, within the past year, Pfizer and Ionis pharmaceuticals announced that they were discontinuing the clinical development program for vupanorsen. And this decision was based on analysis of data from a phase II B study evaluating vupanorsen in patients with dyslipidemia.

Results showed that treatment was associated with statistically significant reductions in triglycerides, non-HDL, and ANGPTL3, however the non-HDL and triglyceride reductions were not large enough to support continuation of the program, which was being conducted with the aims of being able to lower cardiovascular risk and effectively treat severe hypertriglyceridemia. Also, treatment was found to be associated with dose-dependent elevations of liver fat, as well as ALT and EST levels. So unfortunately, it looks like this may not end up being a viable option, in terms of future treatment options. But with the number of ongoing trials, I am hopeful that there will be additional options to help these patients moving forward.

So finally, I wanted to go back to the case that I initially presented. So for the question of what type of lipodystrophy does this patient have? So as a reminder, she had onset of her symptoms early on in infancy, with diffuse loss of adipose tissue throughout, with sparing of palms and soles-- which, if you'll remember from my previous slides, this is consistent with a diagnosis of congenital generalized lipodystrophy. She did have genetic testing, which showed mutations in the AGPAT2 gene, which again, is one of the most common mutations seen in congenital generalized lipodystrophy.

When we left off, she was having worsened hypertriglyceridemia and worsened glycemic control, despite escalating doses of insulin. At that time, she was actually enrolled in the NIH clinical trial and was started on metreleptin. Within that year, her A1C improved to 5.1 and she was able to come off all of her insulin. Other markers, including triglycerides and LDL, also improved on metreleptin.

At the time that I saw her, she was 24 years old, and remained on metreleptin therapy and was doing quite well. She did recently have a brief gap off of therapy due to some issues with insurance and costs of the medication, which did lead to some worsening of her numbers. But since getting back on it, she was tolerating it and responding well to the treatment.

So to summarize, it's important to consider the diagnosis of lipodystrophy when you're seeing a patient and notice loss of subcutaneous body fat. They may appear more muscular and associated with significant insulin resistance, hypertriglyceridemia, or hepatomegaly with steatosis, and especially if they're not responding as well to traditional treatments. Remember that lipodystrophy is a clinical diagnosis. And in these patients, it's important to monitor for and manage any metabolic abnormalities that they may have, including diabetes, high triglycerides, or proteinuria. For patients with generalized lipodystrophy, remember that metreleptin is FDA-approved for treatment of these patients.

And remember that the US label comes with two box warnings regarding neutralizing antibodies, which can be associated with loss of effectiveness of the medication, as well as severe infection. And then T-cell lymphoma as a second box warning. Also remember to look for and consider ongoing clinical trials, especially in those with partial lipodystrophy for whom metreleptin has not been approved for treatment. So these are my references. And with that, I thank you for your attention.