A. KIM RITCHEY: Well, good morning, all. Thank you for getting up early to listen to grand rounds about hot topics in pediatric hematology oncology. So the topics that I'm going to talk about today will provide new information, I think exciting information, and in some cases probably even groundbreaking information on the management of some old diseases.

The areas that I'm going to talk about are hemophilia treatment, relapse leukemia, and stroke and sickle cell disease. So if we first start with hemophilia, you all maybe know that Queen Victoria was the head of the royal families of Europe. And she was actually the first to probably carry the hemophilia gene. And you can see that the hemophilia was spread throughout the royal houses of Europe.

The most interesting to me is Alexei. He is the Tsarevich of the Russian royal family, the Romanovs. So there's Alexei down there.

So Alexei was one of the royalties that had hemophilia. There's a very fascinating book that turned into a movie called *Nicholas in Alexandria* by Robert Massie-- Robert Massie's son, by the way, had hemophilia, which got him interested in the Romanovs-- that describes what life was like with hemophilia before treatment. And it was really, really amazing to see how devastating this could be. It described how Alexei would have a bleed into a joint. He would be pulled around in the wagon for weeks to months on, finally get to walk again, and because of his irritated joint in very short order would have another bleed.

So the family was basically beside themselves about how to manage this. So along comes Rasputin. Rasputin was actually a peasant, but he was a mystic. And somehow, he got into the good graces of the Romanov family, especially Alexandra.

And what he did was he went and helped Alexei through these crises. And we think the way he did it was through hypnosis. The scientific doctors at the time all pooh poohed what he did, but in actual fact, that might have been the best thing to do back in those days where we had no treatment. Well, hopefully we've come along a little bit further than the days of Rasputin.

So indeed, we've known about the history of hemophilia many, many years before the Romanovs. That was first described in the Babylonian Talmud. In early 1800s, it was recognized as the bleeder's disease. Then in 1839, the first term hemophilia was used. It took to the 1900s to define the deficiency of the anti hemophiliac factor that was the cause of hemophilia.

And then in 1947, Brinkhous and Quick defined the role of factor VIII and the deficiency of factor VIII in hemophilia. And also that year and subsequent years, it was clear that it wasn't just one factor that was deficient that led to hemophilia. There were other factors as well.

So we have hemophilia A, factor VIII deficiency, hemophelia B, factor IX deficiency, and others. So just some basics again-- hemophilia A is the most classic type of philia. 85% of patients with hemophilia have that. Hemophilia B or Christmas disease, about 15% of patients-- that's factor XI deficiency.

When we talk about the severity of hemophilia, it's defined by the degree of deficiency. So in patients that have severe deficiency as defined by undetectable, less than 1% factor activity, they compromise the majority of cases of patients with hemophilia. And these are the kids that have spontaneous bleeds.

If you have moderate or mild disease, you have some factor around that ameliorates the clinical picture. And usually those kids will get bleeds only with trauma. They're the minor portion of the hemophilia population. And we're going to focus on this severe population in our discussion.

So what are the clinical manifestations of hemophilia? Well, here's a little baby that I bet if we presented to the emergency department people would say probably was abused. But indeed this is a little baby that had hemophilia.

So we worry about trauma in these little kids no matter what and we worry particularly about head trauma. Thankfully, this little boy didn't have severe head trauma. He just had bad periorbital hemorrhages. But intracranial bleeding is a potential fatal complication of hemophilia.

But the biggest complication in hemophilia is that of joint bleeding or hemophilic arthroscopy. Basically, what happens is that there's bleeding into a joint. The blood increases until it's basically stopped by the confinements of the joint capsule, and it slowly resorbs over time, leaving the synovial lining of the joint very irritated. And this leads to recurrent bleeding until ultimately over time, you have the synovial lining basically disappearing. And eventually, you have bone on bone in terms of any joint interaction.

So in the days before treatment, this recurrent hemophiliac bleeding would lead to crippling of almost all of the patients with severe hemophilia. Most of the boys, by the time they're in their second decade, certainly their third decade, were on crutches and in wheelchairs. And the management of hemophilia has been an attempt to try and stem that tide. The first attempt to treat him was a transfusion of blood back in the 1840s.

But it wasn't until the 1900s where we knew a little bit more about the cause of this that more rational treatment was given, including transfusion of plasma, which did work. But the problem is there's a very small amount of factor in a large amount of plasma. So to get another factor to help stop the bleeding, you had to basically flood the patient with plasma.

Then comes along Cohen, who is able to fractionate the different portions of plasma and fractionate the different protein portions. And then Judith Graham Pool figured out how to take the cone fraction that had the factor VIII in it and actually use the cryoprecipitate. This is the portion of the frozen plasma that as you warm up the plasma remains as a cryoprecipitate. And that had a very high concentration of factor VIII in it.

Well, cryoprecipitate, even in my lifetime, was a commonly used treatment in management of hemophilia, and it really allowed patients with hemophilia a lot more freedom. Prior to cryoprecipitate, they always had to come to the emergency department to have their IV started and factor given or plasma given. But with cryoprecipitate, we trained parents to start IVs, and they put their cryoprecipitate in their freezer, and they were able to treat their kids at home.

But the real big breakthrough came with lyophilise concentrates. That's when the companies took 10,000 to 20,000 donors and they took the plasma and they made the cryoprecipitate. And then they lyophilised it into one little small vial.

So this small vial didn't have to be frozen. It could be put in your backpack and you can go on a trip if you wanted as long as you knew how to infuse it. So this again opened up the lives of boys and men with hemophilia. And in the 70s, it was really kind of a heady time for patients with hemophilia because of the freedom that this new factor has allowed.

When do kids bleed? Well, their first bleed on average is around nine months. It's not usually a joint bleed. It's some other type of bleed. And then if you look at when the first joint bleed is, on average, it's around 16 months, but there's a lot of variability there.

The number of joint plays per year in the natural history of this is somewhere between 30 and 35. So you do the math. That means a boy needs to have two to three infusions, sometimes four infusions, every single month.

And the standard of care in the United States up until 2007 was on demand therapy. That is, when a kid has a bleed, you take him to the doctor or you infuse the factor at home, and that stops the bleeding. That's on demand therapy therapy.

And on demand therapy did a lot to improve the lives of boys with hemophilia. They didn't always move progressively down that hemophiliac arthropathy course that I just told you about. However, it still did not prevent damage, and there were still a lot of boys that had problem into their second and third decade.

Well, unfortunately, we have the tragedy of the 1980s. This is when HIV was discovered, and it was actually because of a hemophilia patient who developed HIV that it was clear that this was from something in the blood. And basically, every single patient with severe hemophilia that will receive the lyophilised factor products-- because remember, it was from 10,000 to 20,000 donors, some of whom had HIV. Basically, everyone got HIV, and the whole population of hemophilia was decimated.

But it led to some great changes in factor product. One was the viral inactivation that was performed on these products-- pasteurization being one product, but there were others. And these inactivation steps basically freed the plasma derived products of HIV, hepatitis B, and hepatitis C, the big three viruses that were a big problem in patients with hemophilia.

So since 1987, in any plasma derived product, there's never been a case of transmitted HIV, hepatitis B, or hepatitis C.

Then comes in the late 80s and early 90s the recombinant factor VIIIs and factor IXs. This again was a great breakthrough because you didn't have to use human blood product to make the factor IX. You could use DNA technology, recombinant DNA technology, to make the factor.

Maybe you used a little bit of albumin in some of the products, but there are even products that don't even use any albumin. So they're perfectly free of any human blood product. So this was, again, took another worry off the parents of kids with hemophilia.

Now the half life of factor VIII is only 8 to 12 hours, the half life of factor IX a little bit longer, between 18 and 24 hours But it's highly variable. So what you can see is that this is a treatment that is effective and we now have a way to deliver it at home. But it's going to be something that continues to be a problem as long as you have to start IVs and treat these little kids.

Well, then comes the Swedish experience where they experimented with a program of prophylaxis. So the whole concept here is to take boys with hemophilia very early in life, usually between the ages of 1 and 3, and turn them from a severe hemophilia patient to a moderate or mild patient with hemophilia. So you give them factor three times a week if they had factor VIII deficiency, hemophilia, or twice a week if they had factor IX deficiency. And this would maintain their factor level high enough so that they only had bleeds with trauma.

So they presented data that was a very, very conclusive. I think everyone was convinced that if you could maintain prophylaxis, you could totally prevent damage to the joints. You can keep kids in school every single day, keep people at work. It was a very effective program.

It really, however, came to the forefront in the United States after there was a randomized trial by Marilyn Manco-Johnson between standard demand therapy and prophylactic therapy which clearly showed that prophylactic therapy prevented joint damage. So we now use prophylaxis as our standard approach. We usually start it between the ages of 1 and 3.

If you've got hemophilia, it requires factor VIII infusions three times a week, hemophilia A. If you've got hemophilia B, it requires infusions twice a week. So again, this is all good improvement, but you know, it's pretty tough for the kids.

So you can imagine a one-year-old, a little chubby one-year-old, that you have to start an IV three times a week to give them factor-- not an easy task. OK, so how about ports? Yeah, OK, you put in metaports, which we did a lot of.

And what we found was that between 25% and 50% of boys with hemophilia develop upper venous clots. And there was a high incidence of infections. We had a kid develop bacterial endocarditis. So that's not a good solution either.

I wonder if there's a better way to do it? Perhaps if we extended the half life of factor VIII and factor IX to decrease the infusions, that might help. And indeed, that's what was new, these extended half life projects. In the last two years, there have been release of two or three, maybe four different extended half life products for factor IX as well as for factor VIII.

And I'll just talk about one product, the ALPROLIX product, which combines the factor IX, and it fuses it with the FC portion of IGG. So this is the fusion product of factor IX. The concept here is that the combined product is engulfed by the cell as it's going to be degradated. But it attaches to a naturally occurring fetal neonatal antigen.

So it attaches to that neonatal antigen, and that neonatal antigen keeps it from lysosomal degradation like other proteins and shunts it toward the surface of the cell. And then it is released back into the blood to continue to do its work. So this is a study from-- a recently published study looking at the duration of factor IX activity with recombinant factor-- that is, the standard plasma derived factor-- versus the new fusion product, OK?

And what you can see is that with the new fusion product, there's been a two-fold increase in period of time over 3%. This is what you want to achieve if you want to turn a kid from severe into a moderate to mild hemophilia. This is the factor VII activity-- so different protein, same concept.

And you can see this is a standard factor VIII molecule. This is the fusion factor VIII molecule. And this is at a lower dose and this is at a higher dose. But the bottom line is that you have an increase in half life of 1 and 1/2 to 1.7 times-- not as good as factor IX, but improved.

So what has been the impact of extended half life products? Well, there's decreased frequency of infusions for prophylaxis. With hemophilia B, we only need to do it once a week. And I have a patient that's on it every 10 days and does very nicely.

Hemophilia A, twice a week. It decreases treatment fatigue. We have to use fewer IVs and required to use fewer metaports in these little kids. Some people who are on on-demand therapy now are converted to prophylaxis because they can achieve it without too many more infusions. And you can achieve higher trough levels and thus decrease the breakthrough bleeding that we can see even when using this prophylaxis program. So this has been a very important advance in the management of boys of hemophilia.

But I really want to-- but I want to tell you now about one that's to me even more exciting. This is a new bi specific antibody that mimics factor VIII. It's called emicizumab. This was published just like three weeks ago in the *New England Journal.*

So the concept here-- well, first of all, background. So this is your factor eight molecule. It connects with the factor IXa and factor Xa to form this complex, which then leads to thrombin information and the clotting process.

Well, this new bi specific antibody basically mimics the function of factor VIII. It hooks up the factor IX and the factor X into this product. So the question is, does it achieve what the naturally occurring product does?

Well, this article published looked at three cohorts of patients with escalating doses of this emicizumab. It was a 12 week study. I want to point out that it was a weekly dose, and it was subcutaneous, not IV, OK?

So if we look at the first panel, what we can see is that there's an increase in the plasma level of emicizumab based on the dose, goes up. Every single patient that was receiving emicizumab had their APTT corrected. And then all of them develop thrombin activity.

Even perhaps more impressive is the effect on bleeding. This is the annualized bleeding rate, red before they gave the emicizumab and then afterwards in blue. You can see here that it reduced it almost to zero in every single cohort.

I didn't have a chance to talk much about inhibitors in patients with hemophilia. That's one of the feared complications of hemophilia. But this works in boys with inhibitors as well.

So this is a very exciting product and I really think could change the whole treatment of hemophilia. It's still-- this is a phase one phase two study. It still has to undergo a lot more research before it can get to market, but this is very exciting.

OK, let me move to leukemia. I think most of the audience knows that leukemia is the most common cause of cancer in childhood followed by CMS tumors, neuroblastoma, Wilms tumor, non-Hodgkin's lymphoma, and Hodgkin's disease. However, if we wanted to put relapse leukemia into this list, it's number four.

So relapse leukemia is a big problem in children with ALL. Now we've done an amazing job over the last 50 years or so in treating children with ALL. Back in 1950, zero children with ALL survived. Now through a series of discoveries as well as treatment trials in CCG in the children's oncology group, we've increased the survival of children with ALL to about 80% to 90%. This is I think one of the great medical stories in the last century.

But there are problems. This is an overall survival probability for patients of different ages. So if we look at infants less than a year of age you can see still about half of them relapse. The age 1 to 9 is the best group.

They do quite well. But the older you get, the more likely you are to have a relapse. So relapse is still a significant problem.

What are prognostic factors if you do relapse? The time of relapse is probably one of the more important ones. If you have an early-- by that, I mean on treatment relapse-- that's a poor prognosis.

If it's after the end of treatment, it's still not good, but it's better. The site of relapse makes a difference. [INAUDIBLE] relapse worse than any other site. T-cell relapses are always worse than b-cell relapses, and there are other factors that affect prognosis, including genetics and response to therapy as measured by minimal residual disease.

So how do we treat relapses? We use pre-induction chemotherapy to get them into remission, giving them post remission chemotherapy. And transplantation is a standard treatment for all patients that have early relapse of ALL. Either either an [INAUDIBLE] transplant or a matched unrelated donor transplant, that is standard. If it's a late relapse, we have discussions about whether chemotherapy may be as effective as transplantation.

So how do we do in the treatment of children with acute lymphoblastic leukemia? Well, this was a study published in 1998 by Paul Gannon that looked at the six year survival by time inside the relapse of kids with ALL. Here's the time.

So 0 to 17, if they relapsed in the marrow, 6% six year survival. And if it was combined with CNS or [INAUDIBLE], 9%. Again, this is on treatment-- 11%. A little bit better if it's off treatment, you know, but not even 50% better if it's an extramedullary relapse.

Well, we've tried to improve therapy. What did a decade do? So this is a study of COG trials of five year survival.

And if you look here, 0 to 17 months relapse, we're up to a big 12%, OK? So not much better later on. Again, a little bit better in all of these other categories, but still a significant problem. We need help for kids with relapsed ALL.

I'm going to show you this one study because it is actually the best outcome for patients with relapsed ALL. It was the UK study that was published in 2010. All comers of ALL relapse entered on this trial, and they were randomized to two drugs-- mitoxantrone and ibarubicin.

And the point that I want to make here is even though mitoxantrone beat out ibarubicin, it still wasn't great, you know? It was still 64% with the mitoxantrone progression free survival, and survival is about the same. So if you combine the two, we still have an overall survival of about 50%.

So we still have problems. Even if, and especially if, you relapse after a transplantation, again, your chances of doing well are extremely poor. They're not zero, but they're extremely poor even after-- if you relapse after transplant.

Thus enters this new treatment called CAR T-cells, or Chimeric Antigen Receptor T-cells. The whole concept of this is taking the endogenous T-cells of the patient and reprogramming them-- reprogramming them so that they're specific for the CD19 that is on the surface of almost every ALL cell. And the concept here is that a lentiviral vector takes the DNA of this anti-CD19 antibody.

It's placed into the cell. Then the cell will express the anti-CD19, and you now have basically a little guided mini bomb. And when it sees the CD19 positive cell, it will attach to it and kill that cell.

So this is the concept of CAR T-cell therapy. We know that you have to have fairly high proliferation of the cells for it to work, and we have to have an extended period of time where they persist to make it work. And those are areas that are being studied.

Now it's not a simple process, and I just wanted to show in this how it takes place. You take the patient that has leukemia relapse, and you do a leukapheresis. That is, you get their T-cells. And then you put their T-cells-- and then you put the lentiviral vector with the anti-CD19 into those cells.

You allow them to grow for a period of time, and then you separate out the T-cells from the beads that they grow on. And then you give the patient chemotherapy. And then you get back the T-cells. So this is not a quick process. This takes weeks to months.

OK, so I want to talk about two papers where this has been used in children with relapsed ALL. This was the very first study in ALL by Steve Grupp and colleagues at CHOP. There were other studies of CAR T-cells, but this was the first for relapsed ALL.

They just reported on two children who had relapsed ALL and refractory pre-B-cell ALL. The first was a seven-year-old who had been diagnosed two years earlier, had relapsed 17 months-- remember, that's an early relapse-- 17 months after diagnosis, had a second complete remission, but then relapsed four months later and then was unresponsive to further chemotherapy. This child in modern day treatment had no hope of survival.

So the CAR T-cells were given to this patient. On day four of five, the patient developed very high fever, respiratory and cardiovascular compromise, had to be put on a ventilator, but with the use of anti-cytokine agents, etanercept and tocilizumab, had rapid resolution of the symptoms. So this is felt to be a cytokine storm or basically comparable to macrophage activation syndrome or cytokine release syndrome.

Patient two was 10 years old, had had a second relapse after having had a bone marrow transplant-- again, no chance for survival to any extent. This kid also had fever a little bit later, developed confusion, felt to be a part of the cytokine release syndrome that resolves spontaneously without any interruption, without any intervention. So this is a complex slide, but it talks about-- it shows patient one and patient two.

This is the fever of that first patient. And this is LDH, which is basically a surrogate for tumor killing. And this is the second patient, a similar curve and these are all the measurements of the cytokines that took place.

You can see the cytokines peaked, including IL 6 in particular, about the same time that you develop the fever and the other symptoms. And this is the white count ANC and ALC. You can see that it went up a little bit more slowly in the second patient.

So the outcome of these two patients-- first all CD19 positive cells, all blast cells, were eliminated from the marrow of these patients that were, remember, refractory. Patient one actually had a remission for nine months and had blood counts that were normal for 11 months. The patient two did have a clinical relapse two months after the CAR T-cell infusion.

Well, Shannon Maude then updated the experience at CHOP by reporting on 25 children and five adults receiving the CAR T-cells. 88% of these patients were in their second or greater relapse. But the results of this study confirmed the smaller study-- 90% of the patients had a complete remission after the infusion of the CAR T-cells.

All of the patients had some evidence of cytokine release syndrome, but only about a quarter had severe cytokine release syndrome that was managed effectively by tocilizumab. This is her probability of event-free survival and overall survival in six months. And what you can see is that the survival rate at six months is 67% and survival is 78%-- again, pretty impressive.

She also pointed out that the correlates to the cytokine release syndrome depended a lot on the levels of interleukin 6. So if you had high levels of interleukin 6, you had a higher chance of having cytokine release syndrome. If you have a higher disease burden, you have a higher chance of cytokine release syndrome. So these were things that could be theoretically manageable.

So I wanted to give you the 2015 update. This was reported just in December by Steve Grupp, who reported all of the patients that had been in this pediatric ALL phase one and two study now up to 59 patients. These again were in second or greater relapse or refractory.

2/3 of these patients had relapsed after stem cell transplant. 93% of this population had a complete remission one month after receiving the CAR T-cells. 18 patients remained in remission greater than a year, 13 without any further therapy. Median follow-up was 12 months with a range of 1 to 43 months. There were relapses, both CD19 positive and CD19 negative, and there were patients that went on to receive stem cell transplant or donor lymphocyte infusions.

So this is the update of those relapse free and overall survival curves. And what you can see is the six month relapse free survival curve in Steve's update was 76% six months, and 12 months, it was 55%. Overall survival is now at 79%.

So there are toxicities, as you saw with this treatment. You get b-cell aplasia when you give this treatment. This is observed in basically all of the patients who had responded. And you have to give them IVIG replacement, but it's manageable.

Cytokine release syndrome is seen in 88% of the patients, but it's reversible. You can treat it with anti-IL6 therapy, and you don't even need to use that if you can control the disease burden before you give them the CAR T-cells. Macrophage activation syndrome is the worst manifestation of the cytokine release syndrome, and you have to be on the alert for that. There also was neurotoxicity seen with this treatment. 5% or so has seizures. Some people had confusion afasia that tended to be transient.

So what's the future for CAR T-cells? Well, we in the Children's Oncology Group feel it does indeed have a future. And we're going to use it in patients that have CD19 positive b-cell ALL.

And what we're thinking about doing it is using it up front in newly diagnosed patients. But we're going to take those patients that have the highest risk of failure, those that have high risk genetic lesions such as hypodiploidy-- that population has probably a 40% chance of survival with current treatments-- translocation 1719, or a patient that have poor response to therapy. Induction failure do extremely poorly. At the end of consolidation, if you have evidence of the disease, you also do very poorly.

So this is the population we're thinking about incorporating the CAR T-cell therapy, and of course if you've got an early bone marrow relapse for all the reasons that I showed you. So this is an exciting therapy, and I think it will be spreading throughout the country. Indeed, it already is.

OK, moving to a new subject, I think you all recognize the sickled cell on this electron microscope. So just as a reminder that the pathophysiology of sickle cell disease, we know that there is a point mutation in the betaglobin gene. That point mutation leads to a single amino acid substitution in the betaglobin, switches from glutamic acid to valine.

And this changes the structure of sickle hemoglobin such that under conditions of hypoxia, you get polymerization of the hemoglobin molecule. And when you get polymerization of the hemoglobin molecule, what you get is you get distorted red cells. And those distorted red cells can lead to vaso occlusion. And of course it leads to shortening of the red cell survival.

If we think of the clinical manifestations of sickle cell disease, you can divide it into two broad categories-- those clinical manifestations due to the hemolysis and those due to vaso occlusion. In hemolysis, we see anemia and the complications of anemia, such as aplastic crisis, splenic sequestration, hyperhemolytic crisis, gallstones, pulmonary hypertension, leg ulcers. In the vaso occlusion side, we see pain crises, which are clearly the most common and sometimes extremely severe complications, loss of splenic function, which can lead to increased risk of infection, acute chest syndrome, renal complications, hyposthenuria, papillary necrosis, avascular necrosis, retinopathy. But what I want to focus on today is stroke.

Let me tell you a little bit about the natural history of stroke. This is the age at first stroke. And you can see here, if we look at the different hemoglobinopathies, this is homozygous SS. This is SC, et cetera, et cetera.

So stroke is a problem in the child and adult with homozygous SS. The data that we know, the natural history of this was that 11% of children with hemoglobin SS will have had a stroke by the time they're 20. 24% of patients with sickle cell disease will have had a stroke by the time they were 45.

The highest risk is in the very first decade, with a 333 timed normal risk of that of a normal child and an average age of about seven years. Recurrence was the routine follow up of these patients that had their first stroke untreated. 2/3 of patients that had one stroke went on to get a second stroke, and the second stroke was usually within two to three years. And of course these could be devastating, of course leading to many neurological complications and even leading to death.

So for a long time, it was felt that the strokes in kids with sickle cell disease were due to the small blood vessel problems that you see in other patients with sickle cell disease. And it really was in this landmark study by Stockman and Oski back in 1972 that first documented total occlusion of large vessels within the brain of children with sickle cell disease. So here you see obstruction of the anterior cerebral artery.

Here's another example of obstruction of arterial arteries-- big vessels, not microvessels. And here we have some narrowing of the vessel. So Stockman and Oski brought up the fact that these were large vessel problems, and you could actually document it by doing angiography if you're careful.

Well, what did that lead to? So back in the mid to late 70s, Jean Luscher in Detroit said, why don't we give these kids transfusions? And she actually started a program of transfusions to lower the percentage of hemoglobin S down to less than 30%-- that means you got to give transfusions once a month-- and hopefully will prevent that recurrent, sometimes fatal second stroke.

But before I go to that, just wanted to mention that we now know that the cerebral vasculopathy of patients with sickle cell disease is actually more complicated than we knew many years ago. It's multi-factorial. First of all, you have this stickiness of the red cells sticking into the endothelial surface-- there's hemolysis-- and releasing a lot of pro-inflammatory cytokines. These pro-inflammatory cytokines then will lead to abnormal adhesion of leukocytes to the cell walls. You have platelet adhesion.

You have release of nitric oxide and endothelium one, which cause vasoconstriction. You have enhancement of the smooth muscle surrounding the vessel, ultimately vessel narrowing. And of course that's when you get the inclusion. So this is a multi-factor step.

So can the recurrent stroke be prevented? So Jean Luscher, using transfusions, seemed to be able to do that. Well, in 19-- when was it-- 95, Dr. Pedalo put together data from about six different institutions looking at the retrospective look at whether transfusion programs can prevent second strokes in patients that had a stroke previously.

And you can see in this study that if they had transfusions, they had a much decreased incidence of second strokes compared to those that did not receive transfusions. So we knew that you could prevent the second stroke in the majority of patients. But the next question was, can you prevent that first stroke? Is there a way to keeping that first one from happening?

Well, enter the transcranial Doppler ultrasound. This is a technology which does multiple measurements of flow velocity and cerebral arteries. They directly-- these measurements are directly related to cerebral blood flow and inversely related to the vessel diameter.

And in a landmark study in this area, Adams published this study relating the TCD velocity of cerebral arteries to the risk of stroke. And what you can see here is if the TCD velocity was less than 170 centimeters per second, you basically had no risk of having a stroke. But if it was greater than 200, you had a high risk of having stroke. So how could we use these data to help the patients with sickle cell disease prevent these life altering strokes? Can the first stroke be prevented in patients with abnormal TCD?

Well, you might guess that if you maybe found that population of patients that had abnormal TCDs and gave them transfusions, maybe you could prevent that from happening. So that's the stop one trial. That's where transfusions were-- patients were randomized to receive either transfusions or standard therapy.

Now remember, these were patients that had never had a stroke. They only had an abnormal TCD. So you can see here that the patients that received the transfusions had no increase in stroke or minimal increase in stroke, and they maintained good flow velocity of the TCD because with transfusions, by the way, you can correct the TCD velocity. It returns to normal.

So if you continue this stroke, the transfusions, you can prevent the first stroke in patients with sickle cell disease. This is a 90% risk reduction. So the next question is, OK, we have to give all these transfusions to kids with sickle cell disease. Can we stop the transfusions at some time? It would certainly help the patients and the families.

So this is now the stop two trial, and this is again a randomized study of continuing transfusion or not continuing transfusion. I think the graph speaks for itself. If you stop transfusion, the now normal flow velocity reverted to abnormal, and some children went on to get strokes. So it seems to me we have the following situation-- transfusions have become and are the standard of care for the treatment and prevention of stroke in sickle cell disease.

How long do you need to continue transfusions? I didn't go over a number of cohort studies looking at patients who had had a stroke and stopping transfusions in that population, and it didn't work. They always had recurrence of their stroke.

And I just showed you data from the stop two trial that demonstrated recurrence of the abnormal TCD or the development of stroke if you stopped transfusions. So it seems at the present time that transfusions are going to have to be continued indefinitely in patients with sickle cell disease who had a stroke. But what is the cost of transfusion program?

First of all, it's monthly transfusions. These kids need to come to the hospital. They need to get transfusions, the cost of that.

Most kids need to have a central line because of all of the infusions that they need. We've already talked about risk of infection from metaports and other central venous lines, risk of clots. Children with sickle cell disease have a higher rate of developing alloantibodies found in red cell transfusions.

And indeed, some patients develop so many alloantibodies, you can't transfuse them. Can't find blood to transfuse them. If you continue to give red cells, you're going to iron overload that patient, especially affecting the heart and the liver.

And so that can take its toll. And of course now have to add chelation to try and get rid of that iron from the heart and the liver. So it does indeed take its toll.

So is there a better way? So let me talk a little bit about hydroxyurea. Hydroxyurea is basically a chemotherapy type drug that was used in a number of leukemias, especially in adults. But it has a number of positive effects in patients with sickle cell disease.

First of all, it can lead to an increase in fetal hemoglobin. It can lead to a decrease in white cell and red cell-- decrease in white cells and decrease in reticulocytes. It changes white cell [INAUDIBLE] so they don't stick to the [INAUDIBLE] endothelium as much.

You have release of nitric oxide which helps with vasodilation. So hydroxyurea has been shown in the laboratory to have a number of positive beneficial effects. And this was the first study, maybe the second study, published by Sam Charache back in 1995 about the use of hydroxyurea in adults with sickle cell disease.

The top graph is a result of this randomized trial between placebo and hydroxyurea. And this was the time to first crisis after starting hydroxyurea. And you can see that it was very delayed in patients that had hydroxyurea. And this is the time to the second crisis. And once again, hydroxyurea was beneficial in patients, in adult patients, in decreasing the number of painful crises.

This is a study more recently published. It looked at improve survival in adults receiving hydroxyurea. And you can see once again here that patients who are receiving hydroxyurea have a longer survival. So this has clearly become a standard of care for patients with sickle cell disease that have the more severe courses.

There has not been similar studies, randomized studies, in the older child and adolescent comparing in a placebo controlled fashion the use of hydroxyurea. I think that the sickle cell world has basically accepted that what works in adults will work in the older children. However, there was a lot of concern about safety and utility in babies.

So they designed the so-called baby hug study. The baby hug study was a study of about 200 babies between the ages of nine months and 18 months where they were actually randomized again to either placebo or hydroxyurea. And you can see here that in every measure, whether it's acute chest syndrome-- the hydroxy hydroxyurea is in the red and the placebo in the blue.

Whether it's acute chest syndrome, whether it's pain, whether it's dactylitis, or whether it's need for transfusion, hydroxyurea was better than placebo in every single situation. So nowadays, we start using hydroxyurea with little babies. So it's become a pretty standard of care in the management of patients with sickle cell disease.

What about the management of stroke? That's the topic of this morning. How can we use-- can we use hydroxyurea for the management of stroke and sickle cell disease?

Well I'll report on two studies. One is the TWITCH trial, which stands for the TCD With Transfusions Changing To Hydroxyurea trial. This was published by Russell Ware earlier this year, and it was a randomized non-inferiority trial-- so 121 kids between the ages of 4 and 16, all of whom had abnormal TCDs.

Now patients in this study had received a year of transfusions and had essentially normalized their TCD velocity. And then they were randomized to either continue standard transfusion or alternatively to switch to hydroxyurea and discontinue transfusions. The results are represented in this line from the article.

This is the standard group and the alternative group. I think you can look at it and see that there's not a lot of difference in this mixed statistical model, and this is used in individual data. Bottom line is that there was-- no child had reverted to an abnormal TCD in this particular study. So hydroxyurea, not inferior to continuing transfusions in this particular study.

Then we have this second study that was just published back in April. This is a study from Europe looking at the long term treatment follow-up of children with sickle cell disease monitored with abnormal transcranial Doppler velocities. So they had a large cohort of 309 patients, and they were particularly focused on the 92 patients that had abnormal TCDs. As per the standard of care, if you had an abnormal TCD, you placed them on a transfusion program. And most of the patients went on a transfusion program.

83% of this population had returned to normal of their TCD velocities, which we would expect. Of interest, it was less likely to normalize if you had significant arterial stenosis. So then hydroxyurea was started in about half of these patients after normalization of the TCD after about two to three years of transfusions.

The results are shown here. This is the risk of having reversion to an abnormal TCD or a stroke in this part of the graph. 72% of this population maintained a normal TCD velocity. So there were some patients that had a worsening of their TCD velocity, but clearly majority were able to stop and maintain good TCD flows. They looked at a number of risk factors that perhaps could predict those patients who went on to have abnormal TCDs, and the only one they found of statistical significance were those that had a rise in their reticulocyte count, suggesting an increasing in hemolysis.

So here's where we are with stroke and sickle cell disease today. There's been really a pretty dramatic decrease in morbidity and mortality from stroke in children with sickle cell disease since the 1970s from an 11% incidence down to 1%. That It was the second leading cause of death in the sickle cell population back in the early days.

Now it's rare to have death from a stroke. Transfusions, of course, are the reason for these great statistics, but the treatment comes at a high cost. Hydroxyurea has become a major therapeutic milestone in sickle cell management, and in 2016, it now appears to be a treatment alternative for many patients with stroke so that they no longer have to be on transfusions.