NewSTEPs

A Program of the Association of Public Health Laboratories™

New Conditions Follow Up and Education Webinar

November 2016

Presentations:

Beth Vogel, MS, CGC: Considerations for Implementation of Screening for New Conditions: Follow-up and Education

Sharmini Rogers, MPH, MBBS: Missouri Follow-up for Lysosomal Storage Disorders: Pompe, Gaucher, Fabry, MPS-1 and Krabbe Disorders

Please direct all comments/questions pertaining to this presentation to guisou.zarbalian@aphl.org or 240-485-2736

Guisou Zarbalian:	Hello, Good afternoon everyone. This is Guisou with APHL. Thank you for being on today's New Conditions Workgroup webinar. This is the second installment of our two part webinar series on Considerations for Implementation of New Conditions. Our first webinar took place on October 11th and focused on laboratory conditions and that webinar was recorded and archived on New STEPS website can be accessed from New STEPS website. Today's webinar will focus on follow up and education considerations for implementation of new conditions and will also be recorded and archived on the New STEPS website and we will share that link with you once it's been put on the website. Today's webinar will have two presentations. One from Beth Vogel who will discuss short term follow up and extended short term follow up for Pompe and XALD in New York followed by Sharmini Rogers who will discuss follow up in Missouri for lysosomal storage disorders. I am going to turn it over now to Kim Piper. Kim if you could press star seven on your phone to unmute.
Kim Piper:	Hi . This is Kim.
Guisou Zarbalian:	Hi, great.
Kim Piper:	Hi. I get the pleasure of introducing Beth Vogel. Beth is a board certified genetic counselor and is a research scientist with the New York state newborn screening program. She manages the newborn screening follow up unit and is the project manager for the New York Midatlantic consortium for genetics and newborn screening services. She serves on the APHL New STEPS Screening Committee and prior to joining the newborn screening program in New York, Beth was a pediatric genetic counselor at Albany Medical Center. She received a bachelor of science degree in Psychobiology from the State University of New York at

Binghamton and a master of science degree in medical genetics from Indiana University. Much easier to say Indiana than Binghamton. Thank you, Beth, for agreeing to present today and I will turn it over to you.

Guisou Zarbalian: Beth, if you could hit star seven on your phone you will be able to speak. That will unmute you and then I'm happy to advance your slides for you.

Beth Vogel: Great. Thank you both. Good afternoon everyone. Thank you for listening to today's call. I hope I can give you some valuable information about our experience with implementing follow up and educating both our hospitals and providers about the new conditions. We can go ahead and get started with the next slide please.

First, I'd like to to talk about short term follow up and just share some experience and information about how the cases are being closed for both Pompe disease and ALD. Next slide please.

Reporting the results for these new conditions can depend on whether or not you have DNA information and there's some differences and benefits with each of those different options. If DNA results are available, emphasizing that the screen is still not completely diagnostic can be a struggle. For most providers, once they have a DNA result that identifies a mutation, they consider the baby to be diagnosed, but we still want them to bring the baby in and confirm our finding and follow the usual process. That can sometimes be a challenge. We really still want the baby to have an evaluation. Having the DNA results, however, does help both the newborn screening program and then the specialists on the other end with triaging the referral.

For example, a baby who has two infantile onset Pompe disease mutations, needs to be seen critically and that's a critical result whereas a baby who has two late onset mutations you can be a little bit less frantic about getting them in immediately for those evaluations.

If the DNA results are not available, it's more of a traditional screening model, in terms of the baby needing to come in for the evaluation, but then you have to think about the coordination of DNA testing and potentially prolonging the evaluation time for the family depending on the turnaround time to get those DNA results. You also don't have as much of an ability to triage in those instances. Both of these would work, but there's different things to think about depending on whether you would be able to incorporate DNA into the newborn screening program's algorithm. Next slide please.

For Pompe disease, how do we close the case. In follow up we've reported out the result. The baby has come into the clinic and been evaluated. For us, engaging and communicating with our specialty care center and disease specialists is key. The information on the slide here is what we get back from the specialty care center about the baby's evaluation. Even though we've called

them and we gave them the analyte result, the GAA enzyme activity from the newborn screen and we gave them DNA results, that doesn't give us the full picture of what's going on with that baby. We also want DAA enzyme results that are from leukocytes. Those are from a whole blood sample in our case and results of cardiac studies, so an echo-cardiogram and an EKG is the minimum that the clinicians in New York decided to recommend for Pompe disease evaluation. Urine Pac4, Glc4 studies and a CK can also help look at the likelihood that the baby will have symptoms or have disease as well as whether the baby has symptoms or not. In an infantile onset case, we would expect the baby to have symptoms already even often at the time of referral on evaluation.

Then, if the DNA results were not part of your laboratory algorithm, I'd imagine you would want to get that as well if DNA testing is done. We have DNA testing as part of the laboratory algorithm so we don't request that back from the clinician unless they perform additional studies for example looking for deletion or duplication that's not part of the DNA algorithm in the newborn screening program. Next slide please.

For ALD, closing the case as well, there's been information that we're looking to get back from the specialist about the baby. The very long chain fatty acids should be done on the newborn and potentially the mother and father if applicable depending on the baby. This be specific very long chain fatty acid information we're collecting is a ratio of C26:0 to C22:0, the ratio C24:0 to C22:0 as well as just the straight C26:0. Those three results are collected on the mother, the father, and the newborn depending on the baby's scenario and obviously the father only if the child is a female. We also get results if the baby had plasmalogen studies done. In rare instances, the baby may need to have additional studies like fibroblast studies or NLPA studies, that's where I was mentioning the additional gene testing that can give more information beyond what is done by the sequencing assay in the newborn screening program as well as any clinical findings that could be noted for the baby.

I have a little image here of a cell. You can see that it's depicting a peroxisome and showing the protein which is impacted in newborns with adrenoleukodystrophy, that's the ALDP protein. The red X through it means that that's not working. You should have the very long chain fatty acids that are being broken down and processed by the peroxisome, but instead that can't happen because of mutation in ALDP and you have a buildup of very long chain fatty acids. That's what we're looking for to confirm the diagnosis. The plasmalogen comes into play because the peroxisome actually makes the plasmalogen. In a baby who has too few peroxisomes and has a different condition but that would also be detected by the screening panel as a differential diagnosis, we would see that the plasmalogens in fact low. That can give us more information, if the baby does not have a mutation identified by the newborn screening program, we're also wanting them to have those plasmalogen studies done to evaluate for other peroxisomal disorders so we get a complete picture. Next slide please.

For ALD, as I mentioned, there are some other conditions where a baby could screen positive for ALD and actually not have adrenoleukodystrophy, X-linked adrenoleukodystrophy. They could have a different peroxisomal disorder but the very long chain fatty acids are still elevated because something is going on with the peroxisome where they're not being broken down properly. In a Zellweger spectrum disorder for example, many of the newborns, at least that we've detected with Zellweger spectrum disorder have already been very ill and been in the NICU. For those newborns, we actually get the test result to the NICU as quickly as possible if we see that NICU is checked on our filter paper and the baby is in the NICU, we want to make sure that they're aware that this is a possibility even before the DNA testing is complete.

There's also single enzyme deficiencies that can cause an elevation of very long chain fatty acids. Both of these conditions are pretty rare but they are a possibility in a baby who screens positive for ALD. Your target is adrenoleukodystrophy but like many of the other conditions on the panel, you may see that babies who have these other conditions will screen positive as well. Typically they're values, at least in our experience have been significantly higher than the values of the baby boys with ALD. Next slide please.

I wanted to spend a chunk of my time today talking about extended short term follow up. This may be a new term or one that you may have heard a few times before, but I was asked today to talk a little bit about the impact of variance of unknown significance and pseudo-deficiencies with these conditions. I think that the impact of those can often be, that can put a child into this extended short term follow up framework. I've got some case examples for you and we'll talk through how to think about these new disorders and some of the results that may come out of screening for them. Next slide please.

What constitutes a diagnosis, and this is a slide borrowed from Joe Orthini. We talk about this often here at our program because it comes up all the time. If you have a baby who has low enzyme activity, an elevated substrate, 2 known disease causing mutations, and clinical symptoms or family history, you can say that child definitely without a question has this disease. In newborn screening, we may not have clinical symptom information for that baby, so you're automatically missing this piece of the diagnosis picture and if the baby has 2 known severe mutations, you can maybe say without a doubt "Yes, this baby has the disease." In that case you may see that they have some clinical symptoms already as well, for example with Pompe disease, but if you have a case where you don't have all of this information, it can become a little bit more complex, so I'll go through an example. Next slide please.

This is looking at what we're considering for these new conditions as the different pieces of information help us decide if a baby has a diagnosis. Low enzyme activity applies to Pompe disease, so we see yes that's a piece of helping us decide this baby has the disease, abnormal laboratory tests, for example CPK, which is a muscle enzyme that is done as part of the evaluation, or Glyc4, a urine

test that can give you some more information. If those are abnormal, that gives you another piece of evidence, or for example for ALD we would see elevated very long chain fatty acids, that's another piece of evidence that the baby has the disease. 2 know disease causing, or one for males with ALD, and then clinical symptoms or a family history. This again would be the ideal, that you would see all of these things and be able to say the baby has the disease. Let's take a look at a couple of real life examples of how they would fit into this framework. Next slide please.

This is baby Zoe's example. She has low enzyme activity on follow up testing after a positive newborn screen for Pompe disease, so this is not her newborn screen result, this is her result from a blood sample that was sent off after she went in to see the clinician, and the value was 15.0 with normal being 67.7 to 706.4. She has no abnormal follow up tests, so her Glyc4 in urine and her CPK are both normal. For her genotype, she has one late onset mutation and one infantile mutation, and then has very subtle delay in motor milestones, so one physical therapist thought that she noticed some subtle delays whereas other evaluators thought the baby's motor milestones were normal, so there was some question whether that was there and that it was that subtle that it was being questioned.

For this case with the newborn screening program, do you close this case and say "Okay, the baby has Pompe disease" and would go into a long term follow up process and be followed, or should the baby be monitored over time and to see if she actually develops symptoms and at one point, so baby Zoe would not at this point be treated for Pompe disease. That was the determination after a lot of discussion with the family by the clinician as well as with multiple specialists. That's one example to get you thinking about the types of ambiguity that tend to see. Let's take a look at another example on the next slide please.

Let's look at baby Legume. The baby Legume has low enzyme activity, again on the follow up testing, so 3.8 was his enzyme activity. He also didn't have abnormal follow up tests, and his genotype is one late onset mutation, one variant of unknown significance, and a pseudodeficiency allele, so there can be a genetic change that doesn't cause Pompe disease in and of itself, but that reduces enzyme activity significantly and in combination with other genetic changes could cause Pompe disease, so he has a very complex looking picture genetically in trying to figure out what the impact of those mutations and that pseudodeficiency allele together would be, and also does not have any symptoms on the full evaluations for both motor, low muscle tone, as well as a cardiac evaluation, so would you say that he has a diagnosis of the condition and place him in to start collecting long term follow up data or do you keep his case open and wait to see what happens over time. I think you can start to see why we're calling this section extended long term follow up, that it may not be as clear cut in following his case as it would be for some of the conditions where the baby would have 2 infantile onset mutations and significant cardiomyopathy on evaluation. Next slide please.

I just have one example here for ALD as well. This is baby B. Baby B does not have enzyme information because that doesn't apply for ALD, so we're missing that because it just doesn't exist for this condition. Very long chain fatty acids are elevated. He a mutation but it's never been reported before, which is often the case with ALD. Many of the mutations are specific to that one family. When you look at his testing more, there's no symptoms or family history when he went in to see the clinic. What do we do with baby B? This comes up very frequently and is actually a very common example for ALD. Do we say his he has the diagnosis because his very long chain fatty acids are elevated and he has a mutation? If we can go to the next slide, I've got some thoughts for what all of this means.

The question that we often ask is "Is this extended short term follow up or long term follow up?" And "Can you close these types of cases as disease, and if not when does short term follow up end?" At least from my perspective, short term follow up ends when we have a diagnosis traditionally, but can you say that some of these instances the child has the diagnosis becomes not quite as clear. I like this quote, it was in an article by Dr. Cuomo. It was actually in reference to their implementation for SKID, that's "a centralized tracking and follow up of infants with positive screens by the newborn screening program allows for the evaluation of clinical utility of testing and ultimately leads to screening refinements."

For me, that really rings clear that the information we get from follow up, our number one job is to get the babies into care and to make sure that that connection happens, but then it's also really important that we know what happens with that baby's evaluation so that the lab has that information when looking at their cutoffs and making refinements to the screening process, so deciding whether or not a baby has a diagnosis can actually have some really big implications for the laboratory when they're looking at their assay and trying to improve their false positive rates or make sure the assay's working well and that we can't take that decision to say that a baby has the diagnosis lightly. Next slide please.

There's are really some things just to think about with extended short term follow up. Obviously, I don't have all the answers, but I wanted states that are working to add these conditions, or thinking about it, to be prepared and aware that these circumstances will likely come up for you. Just a few considerations for managing the disease, the consultation and agreement among specialist in the state is really important. The evaluations that are done on the babies could be different from one specialists to the next, but you want to make sure that there's a minimum so that every baby who has a positive screen is getting at least that minimum testing done. That's been really helpful, to have those discussions early on, before screening even starts to say "When a baby is referred to you, what testing are you going to do?" After that initial round of testing it can become quiet different from one baby to next, depending on the clinical picture and that baby's family's decisions and need, but at least having

that first evaluation be consistent is really a useful thing. This is also just a piece of feedback from our specialist in New York State, that care coordination is very time consuming, involves multiple specialists. For Pompe disease you could have genetics, neurology, and cardiology at a minimum involved with a patient. For ADL you would have genetics, neurology, and endocrinology involved with the patient for their disease management. Next slide please.

Just a few special considerations about x linked conditions. In the interest of time, we'll go through this quickly, but just wanted to again give you something to think about as you're planning. Next slide.

This is just to x linked inheritance, so we have a carrier mother. Mom has one copy of her x chromosome of the gene, so let's this is for ALD of the ABCD1 gene that has a mutation, and if she passes that mutation on to her son then he would have ALD. If she passes it on to her daughter, her daughter would be a carrier, but she could also have an unaffected son, an unaffected daughter. This is a limit bit different than an autosomal recessive condition as we've been familiar with with most of the conditions on the newborn screening panel. Next slide please.

There are some implications for carrier females to be identifies, so this would be even if you were only looking at boys with ALD and not screening the females, you would still have to think about carrier females because you're finding moms that are carriers. 10 to 50% of females who have a mutation will develop some neurological symptoms. They're mild and slowly progress and can start in the 30s, but they're still important for those carrier females to be made aware of and to understand that that could develop overtime as they get older. Next slide please.

I wanted to just quickly go through an example of a family. The arrow is pointing to the first boy in the family who screened positive on the newborn screening panel for ALD, and then this is his carrier mother, and when you start to take a family history there's an uncle who's 30 and actually has maybe had some neurological symptoms and not known why and has the later onset form adrenomyeloneuropathy and is now made aware that he has the diagnosis. The aunt to the baby is also identified as a carrier and her son, who's 4 years old and was born before screening started, is now also identified as actually having ALD when it wasn't previously known, so the picture that we're used to of a recessive condition where mostly it's the child and their siblings that are most at risk for the condition, we're now starting to talk about multiple family members that could be impacted by the screen result.

It's important to take a look at the family history and think about that, and in fact we can often, our very first boy who's screened positive for ALD and had a mutation, can look at the mom's name and mom's birthday we collect and saw that he had 5 brothers that hadn't been screened previously. It was like "Really, this is our first one and he has brothers", so working through that family and in the beginning those boys that weren't screened is really important because they

may be very close to the age or over the age when symptoms are likely to start and so it becomes something that is more emergent to work up and evaluate. Next slide please.

Quickly, education, I find that the earlier you can let hospitals know a new condition is coming the better. That may be because we rely a lot on the hospitals to work with us to be kind of that first triage when a baby has an abnormal result and finding the primary care provider and getting the baby into care, so helping them to be prepared as early as possible has been beneficial. For primary care providers, making resources available to them just in time, which just basically means that at the time you're telling them that the baby is being referred having great resources ready for them so that they know what to do, they know what to expect, is really useful. We've tried to reach out to them before screening starts, but because the likelihood they will actually see a baby who has a positive screen or has one of the conditions is less likely that they appreciate that information right at the time when they need it most.

Then for patients and families, having those resources available, having them on the website, families immediately as you all know get online and start reading about it, so they're going to read the worst case scenario of what could happen, so having information that goes through all of the different scenarios can help maybe alleviate their concerns or give them the full picture before they maybe even have a chance to get into that first appointment. Then having those materials in multiple languages is important as well. I've got some examples on my next slide of our brochures. These are on our website, and actually I think New STEPS has them on their website as well. On the left is the front cover of the information for parents on Pompe disease. That's available in multiple languages, and then newborn screening for x linked adrenoleukodystrophy information for parents. We don't have that in a lot of language yet, but we do have different brochures for different test results, so a different brochure for males and females who screen positive and also different brochures based on whether or not a mutation was identified. Next slide please.

Thank you for listening today, and I'm not sure if we're doing questions now. We're at the end, but I'm happy to answer any questions.

Guisou Zarbalian: Thank you so much. That was excellent. We will have time for questions at the end, so please hold your questions everybody. Our next presenter is going to be Sharmini, but I'm going to have Fizza introduce Dr. Sharmini Rogers. Fizza, if you press star 7 on your phone to unmute, please.

Fizza Majid: Can you hear me?

Guisou Zarbalian: Yes.

Fizza Majid: Okay. Hi this is Fizza. It's my pleasure to introduce Dr. Sharmini Rogers. Dr. Rogers is the chief of the Bureau of Genetics and Healthy Childhood in the

Missouri Department of Health and Senior Services. She has overall responsibility of the Missouri newborn screening programs, genetic programs such as cystic fibrosis, hemophilia, sickle cell, [inaudible 00:29:04] program for individuals identified with metabolic disorders. She represents the department as the state's genetic coordinator and enrolled as the chair of the steering committee for the Heartland Regional Genetics and Newborn Screening Collaborative. In addition to the genetics programs, Dr. Rogers also has responsibility for programs that promotes healthy pregnancies, healthy babies, and children. Having a broad scope of programs under her supervision enables her to promote knowledge genetics and health throughout the lifecycle. Thank you Dr. Rogers for agreeing to present your experiences to us. Dr. Rogers?

Guisou Zarbalian: Sharmini, if you can press star 7 on your phone to unmute, and then I will advance your slides for you.

Sharmini Rogers: Thank you very much for the introduction and I'm very glad that Beth went before me because I think she set the stage very well and I probably could go through my slides a little faster. I'm very glad to be here this afternoon. I thought ... thank you. I thought I would just give you a broad overview of our implementation steps, and what I really want to focus on here is that we created a task force and these were individuals from our 4 genetics centers represented from the state lab and the follow up program here, and we also have a consumer, a patient with Fabry. This task force helped develop basic follow up guidelines, which suggested that tests in labs need to be done. As you all know, all guidelines have to be dynamic, and so as we started screening, and we changed this several times over with the experience that we've had. We also met in person and by phone frequently to review our cases, testing our methodology, our cut off, our division scheme, then any other concerns that we wanted to talk about. We had already established contracts with the 4 genetics centers in Missouri, so all we had to do was to renew those contracts and provide that addition funding for the lysosomal storage follow up. Next slide please.

I just wanted to show you all a map of Missouri and how we are divided into 3 regions and how the 4 hospitals [inaudible 00:31:47] Mercy takes whole of region 1, University of Missouri University Hospital has region 2, and the 2 hospitals St. Louis Children and Cardinal Glennon in region 3. Even though it looks like region 2 is very large with one hospital, most [inaudible 00:32:09] in both urban areas, St. Louis and Kansas City. The abnormal screening results are sent to our designated genetic centers to the primary care providers and the follow up staff. As soon as they get the results, the referral centers usually contact the primary physician, and if the primary care physician is uncomfortable calling the family, then they will allow the centers to call the family and a plan is developed and appointments are made with genetic disease specialists to do. Sometimes they do the confirmatory labs prior to the baby being seen at the genetics clinic. That's just to say too many appointments. Next slide please.

Since that's already went over with the confirmatory tests, I'll just say that for Pompe the evaluation done typically within 24 hours of receiving a referral. The geneticists bring them in to do the physical exam, genetic counseling, education for family on Pompe. Next slide please.

I just wanted to put this on. I'm not going to go over the test. This is just to say that when we developed our guidelines, we also developed what reporting mechanism that the centers had to report the disorder to the state, and so we decided which test needed to be reported. The centers directly enter this information into our web based data system and close out their cases. Next slide please.

This is basically how we have defined how we would like the cases to be closed out in the data system for Pompe. This slides will be made available so you will have all that information, so I won't go and explain in detail what is recorded, that we decided. Next slide please.

For Gaucher the centers don't see the family within 24 hours. They usually see them as soon as possible, that they can get a slot in for a physical exam and they try to see them as soon as possible because the type 2 and 3 Gaucher usually there are some symptoms that can be detected after birth, so they do like to see them as soon as possible but just not as urgently that they need to come in within 24 hours. Next slide please.

This again is just a report about what we expect the centers to send us. Next slide please.

For Fabry, the confirmatory tests, the centers schedule them in the first available clinic again for evaluation, genetic counseling and education. For males, DNA mutational analysis is only do if the enzyme level is low. However, for females, we do them both simultaneously, and then the mother is also evaluated as well as family members. Next slide please.

That just shows what's reported. Next slide please.

For Hurler, the baby's typically seen in the first available slot, again for evaluation and physical exam, so if a mutation testing reveals a genotype know to be associated with a severe phenotype the patient is usually immediately referred for stem cell transplantation and while that process is going on, ERT's initiated. If the mutation testing revels a genotype known to be associated with a attenuated form then ERT is started immediately. If the mutation testing reveals a genotype which is uninformative, sort of unknown significance where they got the genotype then the patient will require close clinical follow up, but we don't do any treatment. If clinical finding MPS1 are noted within the first year of life and the patient has a severe phenotype, they will be then referred for stem cell. Next slide please.

Again, this is just what's reported. Next slide please.

For Krabbe A Missouri now is screening in house. We used to contract with [inaudible 00:37:12], and so in house we have screening for the GALC enzyme, and a limited DNA analysis, which is just a 30-KB deletion, so as soon as an abnormal screen is determined and is sent to the centers they make contact with the family within 24 for assessment and confirmation as well as getting the [inaudible 00:37:37] levels and DNA sequencing. Next slide please.

Again, this is just for the reporting. Then you can again, and one more time.

As of October 31, 2016 we have screened for almost 4 years and 300,000 births. We've had 151 confirmed infants with lysosomal storage disorders, however 25 of those with genotype of unknown significance or genotype of unknown onset in Missouri. Beth earlier gave examples of the extended short term follow up where cases may not be closed. We discussed, and we decided that we would close our cases for short term follow up, but the centers of course continuing to follow these kids up and provide this information on a frequent basis, so for the unknown significance basically if we don't know the, if it's a variant of unknown significance or if it's a late onset mutation or there are 2 late mutations we consider them unknown significance, no not that. If it's an unknown onset, it's disease causing but we do not know when it will manifest. If it's an unknown significance, we know they have the disease causing mutation, but we just don't know when it will manifest or if it will manifest. Next slide please.

For Pompe, we have 40 confirmed positives for Pompe, and it appears that there are slightly more males than females and most we've seen in the white non-Hispanic population. We've had 8 infantiles with 7 CRIM positive and 1 CRIM negative, all on ERT and all are doing well, so we have 24 closed out as the late onset mutations and they are being followed up by the genetic centers, but they are not on treatments. We have 6 closed out as the genotype of unknown significance and 2 with unknown onset. Pompe. We've had quite a few pseudo-deficiencies, and I'll talk about that a little later down. Next slide please.

For Gaucher, we have 5 that have been confirmed positive and closed out as having Gaucher. 1 was closed out with a genotype of unknown significance. Of the 4, 3 were non non-neuropathic, 2 were white males with homozygous N37OS, and 1 of them was from the Amish population, and 1 girl who is American Indian with N409S and the N483P, so then we have 1 neuropathic case, which was a white female. Just to tell you a little bit about the follow up, the first type 1 Gaucher disease that we had, that child is now 16 months, has [inaudible 00:41:14], we initiated therapy. The second case was diagnosed with Gaucher type 3 and they were symptomatic at birth, had [inaudible 00:41:26] and some neurological abnormalities and started on ERT. The other 2 cases are both type 1 and not currently on any treatment. Next slide please.

Sorry, my screen just shut down so I can't see what slide it is. Okay for Fabry yeah.

Guisou Zarbalian: Yes, go ahead.

Sharmini Rogers: For Fabry we have 86 cases closed as Fabry, 81 males and we had 6 females. The majority of the population are white and non-Hispanic, and we had 5 that we closed out with genotype of unknown significance. What we have found is that the PA143T allele is very common. It's seen in 59% of our cases, and we have identified multiple family with Fabry, and some actually we they got those family members in were symptomatic but had never been diagnosed previously. The one identified as classical Fabry we are able to do that because we knew the mother had classical Fabry and the mom and baby had the same mutation and she'd been treated by one of the centers. The A143T has been associated with non-classical Fabry disease and a lot of questions have been raised here in whether this mutation is pathogenic because we have such a large prevalence in Missouri. The centers do not consider females with the Fabry mutation as carriers. We class them as having the Fabry disease, since the females with mutations have all shown symptoms. Next slide please.

We have 4 cases of Hurlers confirmed and 2 have been closed out as MPS1 as severe, 1 male and 1 female, and 2 we closed out as genotype of unknown significance. The first case of MPS1 had multiple abnormalities and died with complications after transplantation. The second one was successfully transplanted and continues to be doing very well. Of course, Hurler we have quite a few pseudo-deficiencies I'll address at a later point. Next slide please.

For Krabbe A we have 11 confirmed positives and after 4 years we actually have 1 that we closed out as infantile onset. The child was just diagnosed as the infantile Krabbe A and is now being treated at Duke. We have 4 closed out as unknown onset and 6 closed out as genotype of unknown significance. All are being followed up either by the PCP or neurology and 1 of the boys who has Krabbe A has Fabry with 198T mutation. Next slide please.

The interesting thing about the pseudos-deficiencies, we close out the pseudos-deficiencies as no follow up needed, but it's just interesting that Pompe we have seen 28 pseudos-deficiencies and it looks like it's slightly more in the males than in females, but we've only seen these pseudos-deficiencies in the Asian population and Pacific Islander. We haven't seen it in any of the other races. Next slide please.

For Hurler, if you can see the minimal pseudos-deficiencies, we have 58 pseudos-deficiencies and again as i said they're all closed out, but the interesting thing to note here is that 51 of them were from African American population and that, yeah I think that's pretty much it. Next slide please.

The way we follow up our late onsets, obviously our 4 contractor centers are doing the follow up, and we all had decided on the guidelines and most of the centers follow a similar pattern, but they don't all do it exactly the same way. Patient are usually, for Pompe, are seen every 6 to 12 months and then if they have low enzyme and a mutation is consistent with the late onset, but if they have low enzymes with 1 infantile mutation then they are seen every 3 months until 18 months and then after that every 6 to 12 months. They all get their baseline tests and when they come back they get their regular reassessments. One of the centers sees the babies yearly and families are counseled that if they're any developmental or motor concerns or respiratory concerns then they should come back, and one of the centers had a family come back prior to their 1 year with those concerns and they tested the urine HEX4. It was a little on the high side of normal, so they a repeated it a month later and it was normal and the CK done was also normal. Next slide please.

For Krabbe A, we didn't detect no late onsets. Variants of unknown significance or onset, we see them on an annual basis and obtain neurology assessments. We also plan to do a psychosine levels. Next slide please.

For the variants of unknown significance we didn't really have any specific guidelines for follow up. The center bases this on markers when they bring them back for assessment, the clinical presentation, and the particular disease that they're following them up. If a child is not symptomatic, they'll just see them back again annually. Next slide please.

For education we developed, we have our newborn screening pamphlet, and we added information in about the newborn screening-

Speaker 6: I just took the [inaudible 00:49:05] lab copy. I'm giving Tina's because we got rid of all of them.

Sharmini Rogers: For our newborn screening pamphlet we just added information on the lysosomal disorders that we were screening, and we also had linked on our website to Baby's First Test that has fact sheets for parents on the disorders. For more information and education to the families we rely on our contracted centers to provide the education and genetic counselling. For the providers we sent out information to our population based pilot screening telling them about what is being screened and how the process was going to be, and we did that same thing again we went live, so when the abnormal results go out to the providers prior to these events the letter would go out to them, to all the submitters, telling them about what was going to occur. The contractors centers also provide grand rounds education about these disorders and about newborn screening as well as they do one on one education to the primary care providers when the cases are referred. Next slide please.

I just put this on to show what we sent prior to implementation when population, when we were doing the population based pilot study, and then next slide please, is the letter that we sent out when we went live. Next slide please.

Basically some of the lessons we learned over the 4 years of screening, that's it's really important to have regular communication between our contracted centers, the lab, and follow up staff, so that we have a place to share our ideas, brainstorm the cases, and make changes to the guidelines. This is all new territory so it has been very helpful for us to be able to do that. This has also allowed us to change our cut offs, adjusted improved guidelines, and then what we really have struggled with is that since this is all new and we don't really have enough information, there's really no clear guidelines on how often these late onset patients should be seen and when actually should they start treatment. There are also no guidelines on specific assessments that needs to be done routinely for these asymptomatic late onset patients. I think our contracted centers are doing an excellent job trying to figure all this out, but it will take a while before we have all these answers, which I think is another reason why it's essential to have long term follow up for this disorder so that we are able to learn from this and get the information and be able to share with everybody else who's beginning to screen.

We also don't really know whether those with the pseudo-deficiency allele and if they have one know diseases mutation are they at risk for developing disease. With that I think I'll stop. Thank you. Before that I would like to acknowledge all the people who gave me all the information and has helped to make our newborn screening program really strong and successful. Thank you.

Guisou Zarbalian: Thank you so much Sharmini. We have a few minutes for questions. We already received the one question in the chat box, which I'm going to read out momentarily, and then if anybody has any additional questions please press star 7 on your phone to unmute. I'm going to start with the question from the Washington State Newborn Screening Program. The question is, what has been your success rate for parents bringing their babies in for follow up appointments for babies with late onset forms or variants with unknown significance? I think the question is for either or for both speakers.

Sharmini Rogers: I'll go first then. I think we've not had a lot of trouble of getting families to come back for their appointments if those that had been diagnosed with late onset and variants. We haven't had many lost to follow up, but I think the one's that were initially diagnosed with Krabbe A with a genotype of unknown significance, that I remember we had some issues with talking to the families, not me the centers, talking to the families and trying to get them to come back, so sometimes they don't come back to the center for their annual check up, but they are coming back to the PCP and the centers that are keeping in contact with the PCP.

Guisou Zarbalian: Thank you Sharmini. Beth, did you have anything to add? I did re-mute Kim and Fizza and Beth. If you would like to answer questions, please hit star seven on your phone.

Beth Vogel: Okay. Can you hear me?

Guisou Zarbalian: Yes.

Beth Vogel: Okay. I don't have any data to answer that question, but anecdotally it can be challenging to get them to come back in. Our longer term experience has been with Krabbe disease, where there's a similar situation that comes up where they need to come back and be followed but are asymptomatic and we've heard from our specialists that it can be challenging to maintain that continued evaluation that's where the amount of time commitment from them related to care coordination and to try and get the family to comply and come in for the follow up studies can be a burden for the centers.

Guisou Zarbalian: Thank you. We have two more questions in the chat box. The first one that came in was for both presenters, did you solicit parents in to it when developing education materials?

Beth Vogel: For ALD, we did. We have a mom who actually advocated for ALD screening in New York state and she worked with us on reviewing the education materials. We have a group that helps us with developing brochures and materials for marketing. They tend to give us insight of the understand-ability as a lay person. We didn't have a specific Pompe parent review those brochures.

Guisou Zarbalian: Thank you.

Sharmini Rogers: This is Sharmini and as I said, we just put a very small blurb about screening for lysosomal storage disorders in our newborn screening pamphlet and we really have used the Baby's First Test for the information. For all our other newborn screening information, if we were to develop anything we usually have a parent in the group to review it. [inaudible 00:57:08] we had a consumer, a patient with Fabry, if we were to develop more, we would definitely consult him. We have definitely consulted parents for all of CTHD [inaudible 00:57:22] and brochure and the other brochure.

Guisou Zarbalian: Thank you. Next question. Can you describe the implementation process of referring patients from their initial hospital newborn screen to their PCP or regional genetic centers? Are you getting issues with PCPs not wanting to do the secondary screening?

Sharmini Rogers: What do you mean by secondary screening?

Guisou Zarbalian: I'm wondering if they're asking about confirmatory testing, maybe.

Sharmini Rogers: We've not had any problems. However, as I said earlier, the way we do it is that the lab sends out the results to the PCP on the newborn screening form and to the designated referral center and to the follow up program. The center will contact the PCP to arrange for what to explain the disorder and develop a plan of what needs to be done. For the most part the PCPs have done the test, send in blood for confirmatory testing if needed and if they were not able to do it then they send the family to the center to get that done. I don't think that's ever been a problem. Here and there of course there's some PCPs that may not follow up. I think for the most part, we've had no problems. Jaime sitting with me here, my newborn screening program manager.

Jaime: I think, generally speaking, it works pretty smoothly because all of our PCPs are relatively used to the process of using our referral centers for consultation and they're really pretty good about following the recommendation that comes from the referral centers.

Guisou Zarbalian: Okay. Thank you. I'm going to go on to the next question. Has there been any issues with insurance covering the long term follow up testing?

Beth Vogel: No. We haven't encountered that as an issue. I think part of in all the information I talked about of if you say they have a diagnosis or not that from the perspective of the specialty care centers, if they have low enzyme activity or elevated very long chain fatty acids for ALD and a mutation, they give them the diagnosis for the purposes of saying that this child has this diagnosis and needs to have this testing done to monitor the disease. I think if they tried to day this child doesn't have the diagnosis that you could maybe have an issue with getting that covered but I have not heard that as an issue in our specific situation.

Sharmini Rogers: Same here. Where we've had issues with getting testing done is if we want to get DNA sequencing, some insurance companies have required justification and some have denied them. We actually have provided the centers with very little money to help with that and we are sort of reevaluating that because getting those confirmatory DNA analysis has been a little challenging.

Guisou Zarbalian: Thank you both. Next question. For ALD, how are you handling polymorphisms and are they being called out?

Beth Vogel: If the very long chain fatty acids are elevated, the baby is referred to the specialty care center for further evaluation regardless of the DNA results. If we saw only polymorphisms, we would let the specialty care center know that that happened. Though, honestly, most of the time, we find a true mutation. If we don't then, except for maybe two cases over the last couple of years, it's been a baby who's been ill and is pretty quickly diagnosed with another peroxisomal disorder. In theory, if we saw only polymorphisms, we would still refer the baby and let the specialty care center know that, but in practice, it really hasn't come up.

Guisou Zarbalian: Thank you. It's three minutes past two o'clock, but before we wrap up, I do want to just take another second to make sure, are there any other questions before we close the webinar? Okay. Hearing none, I want to give a final thank you to our presenters today, Beth and Sharmini, thank you so much for your presentation. This webinar, again, has been recorded and will be distributed once it's posted to the New STEPS website. Thank you everyone for calling in. Thank you. Goodbye.