Jonathan Sebat: Autism Tree's relationship has been really constructive for the scientists here at UCSD, and I'm no exception. I think that I would regard as really one of our key connections to the autism community is really through that the scientists in my lab are actually get to have face-to-face interactions with people with lived experience and actually get direct feedback on our results. I can remember a visit from Otto to my lab where he loved to make me squirm at every turn. We get real frank feedback on the science and on the genetics, and that's great. As you can tell, the title of my talk is a little different. I knew that this was an end of the day talk. I'm also well aware of what is going on in the world right now, with respect to autism. I decided that it was a good time to really describe in the most basic way, what does the science actually tell us about the causes of autism? You've seen many presentations here from neuroscientists who are studying autism genes and making real progress and understanding how that gene influences brain development. But we should step back a little bit and look at the broader data on genes and environment, and actually look at what's going on. You find that you actually have real answers to a lot of the tough questions that people are asking. What some of our representatives in the government are stepping up to the microphone and saying, we actually have answers to those questions. I'm going to start with a bit of a story here. It's a little bit of a triggering story because we're going to talk about vaccines for a minute. But the reason why is because, going back to when I first got into the field in 2005, and the vaccine health scares were front and center, and we had no genes found yet. There was a very common sense reason why people thought that vaccines might be linked to autism. I can describe for you how that works. It was a very common experience in the community that you would have a happy, thriving baby with fairly typical development up to 12 months of age, making eye contact, sitting up and crawling, pulling to stand, responding with joy in their play. Then something happens. That was a very common story where around about 12 months of age, in this case, James does happen to get a routine MMR vaccine, and at the same time, he developed a fever, was fussy. The doctor wasn't worried about it. But a period of failing to meet milestones immediately followed the MMR vaccine. This was a common experience in the community. That was what led people to start looking for answers. There was reason to think, maybe it's the MMR vaccine. The actual symptoms of autism sometimes have their onset at that time. Then, of course, one's experience is often reinforced by talking to other people that have the same experience. It was a very common experience. There were a lot of reasons why common sense would lead you to think that there was a link between MMR and vaccines. Everyone's heard this 100 times. I don't need to go through it again. There have been many studies that have really not shown a link between vaccines and autism. Around about the same time that all of those studies were being done, the genetics field was going very much, into looking at the genetic contributors to autism. What we've learned is that there's a very strong genetic basis, but it's not exclusive. There are contributions from genes and environment, and the genes are really important in helping us to unpack all of that and actually understand how both. The twin studies were very clear. If you had a condition that was 100% genetic, this is what your twin studies would look like. You would have 100% concordance between identical twins and you'd have 50% concordance between siblings or fraternal twins. That's what 100% heritable condition would look like. Now, what would it look like if there was a really strong environmental trigger? If there was a really strong environmental trigger, we have many examples of that. I'm in the Department of Psychiatry, and we study PTSD, which, by definition, involves a strong environmental trigger. It's part of the diagnostic criteria. What you can see is that, this is what it looks like in that case. There is a genetic component, but you only see 30% concordance in twins and only 15% concordance in siblings. That gives you an estimate of about 30-40% genetic. What is autism What did autism look like in our twin studies. Well, autism looks a lot more like Huntington's than PTSD. The identical twins are 70-90% concordant. When one twin meets criteria for autism, there's a very high fraction of the co-twin who also meets criteria for autism, and it drops off as you go into fraternal twins and siblings. These heritage estimates have not really been changing much over time. It was clear that genes are a major factor. What are the genes? I've had multiple people ask me today, what are the genes? The discovery of the genes began in 2007, and it began because we changed the way genetic studies were done. In the past, you had these large efforts that were

collecting lots of cases and lots of controls and genetically analyzing them and looking for differences between the cases and controls. Mike Wiggler and I decided to do something different. We said, Wait a minute, Autism is sporadic much of the time. Why don't we look at the sporadic mutations? And so we started trio-based studies looking for mutations that mom and dad don't carry. Where mom and dad's genome is clean at that place in the genome, and there's a new mutation that's present in the child that's not inherited from mom or dad. We looked at that. That's actually since become the primary strategy for finding autism genes. You may have heard about all the success in the field in finding autism genes. Do you know where those mutations are coming from? They're not coming from mom or dad. The hundreds of autism genes that we know about are de novo mutations. They are new hiccups in the genome that were not inherited from mom or dad. All of this progress that you've been hearing about in genetics, it's not genetics that mom and dad carry. The vast majority of the signal from autism genes is coming from de novo mutation. That would not be true today if not for two things. Number 1, the innovation. Wait a minute. Let's look at sporadic mutation and actually sequence families. That was innovation Number 1, and Number 2 was Jim Simons. When I presented to a room of scientists at Rockefeller University our unpublished data, and Jim Simons was present, he also funded that study. Jim funded our first study, and then I presented this to Jim and a room full of scientists at Rockefeller. It was very clear around the table, there was a consensus that this is a good way to find genes. Let's use this trio-based sequencing to find autism genes. The problem was we didn't have any family collections. It was these big case control studies that were collected for entirely different type of genetic study. There were none. There were very few in the field. Jim Simons stepped up at that meeting, and he said, Let's do it. He has since funded the collection and sequencing of families systematically since 2006 when they started the Simon Simplex collection and all of the 250 autism genes that you've heard about, which were recently at the American Society of Human Genetics meeting this year in Boston, they announced they've now gone 100-250 genes. Those were all found because Jim Simons collected these large family samples and did sequencing of families. All of that signal is coming from the de novo mutations. Why these de novo mutations are so important is because it's very easy to do a genetic study and find a correlation. Then you talk to a vaccine skeptic and they'll say, correlation. I'm not impressed. These are not just correlations. These are not little flags in the genome that are pointing at something, maybe some signal that's there. These are causal mutations. You can take the SYNGAP1 mutation that was found in a kid on the autism spectrum. You can put that into a neuron in a dish and see how the neuron behaves. You can put that into a mouse and look at the development in a mouse. You can also take all of the kids that have that mutation and see if they have specific clinical features. Sure enough you do. The genes that are coming out have very specific clinical presentations. Each gene can be a little part of the autism spectrum, and it's distinguishable from another gene which has a slightly different function. In particular, the synaptic genes are very much more linked to epilepsy, and the other genes that are involved in gene regulation have a different set of clinical features, and you can actually see the differences in the clinic. Then when you put these genes into mice, you can see clear neurodevelopmental effects in the mice. There's no question that these are causal variants. It's very good for science, and I think it's helpful to focus on these genes because these are the ones where the causal evidence is clear, plain as day. It's not all of autism, by the way. This is the ones that we've been able to solve. These are not just risk factors. Many of these genes really are like monogenetic disorders. Now, when you look at them all and you put them all together, they actually tell a story. They're not random functions. They have very specific functions in the brain, like gene regulation, and they're very specifically expressed during fetal brain development. Other signaling pathways in the brain that regulate the growth and differentiation of neurons, and they regulate neurotransmitters in the brain. Even at that point, it was very clear that there's a real biology that the genes are telling you. Also, in the clinical lab, it's starting to have success as well. My colleague, Steve Shearer, did a study where when you combine all of the tools we have for clinical genetic testing now, the diagnostic yield that you get in the autism spectrum is about 30%. If you combine the clinical microarray and the whole-exome sequencing. We can actually try to project if we had all the autism genes, where would we get to? My colleague and

mentor, Mike Wiggler did some of these projections where he's he's basically getting roughly the same diagnostic yield of about 30% today. But they estimate that, the model predicts that we may ultimately get to 387 genes, and that that would give us maybe a 60% diagnostic yield. That in a majority of autism, there would be a meaningful clinical result from clinical testing. It also tells us the most of the genes have not been found yet, although, again the latest set of 250 does, in fact, get us more than halfway there. Now let's get back to the common sense. Now we have real genetic contributors to autism. But what do they teach us about what's going on in development? One of one of the first things that I hear from a vaccine skeptic when I'm talking to them is that, Well, I realize that there's genetics involved, but you weren't there when my kid was developing normally and then started to regress. That doesn't look like a genetic condition. Shouldn't that genetic condition have been obvious from Day 1. Did it suddenly have developed normally and then deteriorated. That was something I very often hear. The genetics is teaching us about that, too. We know about the 30% that you get with regular testing. My colleague, Katrina Williams in Australia, actually specifically collected subjects with developmental regression and did clinical genetic testing on the subjects with developmental regression and got a diagnostic yield of 50%. The individuals with regression actually have a very strong genetic signal and may actually have a higher diagnostic yield from clinical genetic testing. This tells us that there's a strong genetic component here. Again, this is still a relatively small sample. More studies of autistic regression still need to be done, but If you actually look at the genes that Katrina was finding, they are all of the same autism genes that are coming out of the whole-exome studies. You'll see some of these genes on your list that you've seen before, like P2, it's RET syndrome. IQSEC. SCN1A is a very interesting example because it teaches us even more when we go deeper into SCN1A and look at it. We can even look at SCN1A and how they react to vaccines. A study was done to look at the effects of vaccination on someone that has an SCN1A mutation and the parents were not imagining things. Those individuals, shortly after they get the vaccine, they will have a febrile seizure sometimes and there's a peak of seizures, and sometimes it's the first seizure that the baby has ever had. After vaccine, you get a fever and SCN1A Dravet syndrome is prone to febrile seizures. When you have a fever, you're more likely to get a seizure, sometimes the first one and so, basically, it looks like the disease onset correlates perfectly with the vaccine because it does. It does correlate with the vaccine because that's actually a febrile seizure, sometimes the first one. The cause of the encephalopathy is not the shot. If you actually compare long term outcome of Dravet syndrome in people who received the vaccine and who did not, there's no difference in long term outcomes. That one seizure that was one febrile seizure that happened when the shot is just one of many that they're going to have in their lifetime, and there's really not a difference in outcome. It's really the gene is the cause in this particular case. The moral of the story is the parents are not imagining things. There's a very common sense explanation here. It was not a coincidence that the seizures happened right after the vaccine. Some people with high genetic susceptibility to neurological conditions can react in an adverse way to a vaccine, but it's not the vaccine that's causing it. Now the question of the prevalence. It's really hard to increase the prevalence of autism genes in the population. That's not something one can easily do. Justifiably, one would say, come on, how do you explain the rising prevalence with genetics? Now, you can't, at least not entirely. But then again, you have to look at the science of the rising prevalence. If you actually look at the rates, the diagnoses are definitely increasing. The symptoms have not. When you do population level screening of basic ASD symptom scores, those have not been increasing over time. If you just do general population screening of ASD symptom scores, independent of a diagnosis, those prevalences are not increasing. Autism symptoms are not increasing in the population, but the diagnoses definitely are. Another interesting thing is that the rise in prevalence is not attributable to profound autism. The profound autism is not increasing much. It's entirely attributable to the non profound autism. Which is, again, consistent with our diagnostic drift in terms of our better ability to diagnose it. Is it not increasing at all? Again, the genetics allows you to actually unpack this. Again, that last slide that I showed you is not a genetic study. That's just a study of different subsets of the population. If you go into the genetics, you can actually ask this question, is there anything about this that would increase over

time? The autism may or may not be increasing at a population level, but you can look closely at these things, and what you see is that the environmental factors themselves are very closely tied to genes and here are some ways in which real changes in the vulnerable population can happen. For example, the rate of NICU survival in 1980 was 50%. A very large fraction of kids who were admitted to the NICU didn't make it. Now, the fraction of individuals who make it now is 90%. Kids who are admitted to the NICU, the vast majority of them now make it and they didn't make it before and this is a vulnerable group that has a higher real prevalence in the population that has really increased since 1980 and preterm birth and neonatal intensive care is a mix of genes and the environment. My colleague at Rady Children's Hospital, Stephen Kingsmore specializes in clinical genetic studies of the NICU, and ten to 30% of NICU admissions are later determined to be genetic conditions like Down syndrome, Noonan syndrome, skeletal dysplasia, and CHARGE syndrome. These are real genetic conditions that used to not make it that are making it now. There really is a change in the vulnerable population, at least for the ones who spent time in the NICU. That's one little factor, and you note them to be vulnerable from the genetics. The genetics is telling you this is a vulnerable group that's survival rate is increasing. Now, I'll go through one other. MyLab has specialized a little bit in this particular question, and this is also a very interesting one to look at. Parental age has been increasing significantly. Father's age has increased, like four or five years since 1980 on average and so that's another thing and sure as many of you have heard advanced father's age and mother's age are associated with autism, about a 1.5 fold increase for mothers and fathers over 40. Two papers published in 2012, one from MyLab and one from decode showed that in men, new mutations accumulate de novo mutations. Remember the de novo mutations that we were talking about? De novo mutations accumulate in the sperm of men as they age linearly. It's not unusual. It's what all of us do. They're constantly dividing, and they accumulate mutations as they constantly divide at a rate of about one or two new mutations per year. When we saw this, we were that's the parental age effect. It's coming from these de novo mutations. That would be the parental age effect looked like in 2012, maybe it's a simple genetic mechanism. No, it's not simple at all. Again, we have whole genome sequencing now. We're not just looking at de novo mutations, we're looking at everything. Now, if you look at all of the genetic predictors of autism and how they relate to parental age, you see something really interesting. Every single different factor that you can measure is correlated with mom's age or dad's age, every single one of them. They're either de novo mutations that are accumulating in the sperm of men as they age. Those are the two things on the left, de novo loss of function mutations, or they're common variants in the general population that are associated with older being older when you have kids and the biggest one is the polygenic score for education. A genetic predictor of a high level of education predicts that you're going to have children later because people with PhDs wait a little longer before they have kids. Now, you try to dissect this a little bit you can see that parental age is strongly correlated with parents level of education. Wealthier zip codes also have more access to clinical services. Your more affluent, educated, high socioeconomic status zip codes will also get more diagnoses. On top of that, education genes do have a direct influence on autistic traits. That same education polygenic score that I'm talking about is also positively associated with autism. The kids on the autism spectrum, when you look at within families, they have the highest genetically predicted IQ on average. There's genetic predictors of high levels of education are actually directly influencing autism susceptibility in the population. You've got a couple of different things going on here. Again, but the parental age is not the causal factor here. The parental age doesn't cause the education. It's the other way around and so it's a non causal relationship. It's correlation not causation. The de novo mutations are causal relationship. Older waiting longer before dad has kids accumulates real mutations and can cause genes to be disrupted. But the other pathway is not a causal pathway. There's a combination of causal and non causal pathways. The non causal pathway is one in which there's just a correlation between higher levels of education and autism susceptibility. But at the same time, there's a real sprinkling of mutations that also accumulates in the sperm. You have a combination of things. It's really a complex mixture of correlation and causation. As it turns out, the non causal effect is actually significantly stronger than the causal one. It's mostly correlation

not causation and a tiny bit of causation underneath it all and the genetics allows you to unpack it all. Now you can actually see the different components. To summarize, research finds really good evidence for all of the following interplays of genes and the environment. In essence, there is no environmental contributor that's independent of genes. They're always either correlated or linked in some way. You have, for example, the de novo mutations, as I talked about. You also have inherited variants and polygenic risk, and these are genetic influences that influence autistic traits in the population. You also have a variety of environmental factors again, that have been identified, and almost certainly there's real influence, especially the social factors. Social factors are a big one and there's a variety of things that can really influence outcome in a real way, but they're not as big as the genetics. But then, of course, you have things like preterm birth, which is itself a significant risk factor, and contributing to the environmental contributor. Then it can go and again, the preterm birth itself can be a genetic susceptibility. Remember, the NICU, the diagnostic yield in the NICU. There are genes that can contribute to the preterm birth, and that can also contribute to more of an environmental condition, but it can also go the opposite direction. Parental age can introduce genetic changes, and that will increase your risk as well. There's bidirectional interactions between genes and environment. Genetic factors that can determine your environment and environmental influences that can act on DNA. Those two things are both happening at the same time. It's important to study the environmental influences on autistic traits and so it's something that's been in the news. We heard somewhere that they were going to solve the causes of autism by September. I'm not sure why they said that. What was supposed to happen in September was that grants were supposed to be funded to find the causes of autism and in fact, the announcement where I think they announced that Tylenol causes autism, that announcement was the grant announcement of all the funding recipients. That was actually probably the reason for the press conference in the first place. It's important to study these factors, but you have to take it seriously and you can't do. The way epidemiology is happening right now is you just look at observed correlations in the population, and the epidemiologists do their best to try to control for all the little confounders and all that correlation not causation that's going on. But they're just throwing in whatever confounders that are convenient to them that they have available. Not really dissecting it and unpacking it. If you really want to study the environment, we have to really recognize the complexity of it. The environment is vast, constantly changing, hard to measure, and highly intertwined with genetics. New studies of the environment are definitely needed, and incorporating genetics into the design, you need sequencing, you need genotyping of the actual people for which you're measuring an environmental exposure, and you need to look at how they interact. Again, if vaccination had a causal role, the genetics would tell you if that were true or not. If we can measure how much genetic signal there is in group A versus group B. And if a vaccine was causal, you could see that you weren't having a strong genetic signal in the exposed group. There's a there's very simple straightforward ways to test these kinds of things. The genetics can tell you a lot about the environment. They didn't solve the causes of autism by September, but they did do something. This again, all of the attention that autism has been getting in the news in the media and in the government, it's really frustrating. I'm sure a lot of people in this room are really frustrated about everything that's going on, but $50 million is going towards autism research as we speak. This $15 million Autism Data Science Initiative was announced. It was bizarre. It was June. Lilia and I were flying to Australia. To give seminars at Sydney University, and suddenly, this Autism Data Science Initiative was announced with a deadline of less than four weeks. We had to write a $5 million proposal in three weeks, and we did it. I'm proud to say, 200 proposals were submitted, and we were one of 12 groups that were funded in the $50 million thing. If you look at all of the other projects that were funded, this is hardcore science that's being funded. It was the initiative came from some not-so-sound ideas about the causes of autism, but they were put together by real NIH program people at the NI Environmental Health and Safety Institute, and they were sent to real scientists to review. Really good groups that are doing really innovative science are actually funded from this initiative. In the end, it's actually a boost to autism research. Meanwhile, though, there are a lot of grants that were paused or defunded at the same time. I'm one of the lucky ones in that we're still surviving.

There's a lot of scientists out there whose grants have just been paused, and it's just sitting in limbo, and they don't know if it's going to be funded or not. They have employees that they're supposed to be paying. That are accumulating debt in their labs while they wait for the government to come back and then fund the NIH, and then NIH decide whether or not they're going to have the fourth year of their grant actually funded. It's a really tough time in science right now, but there's a silver lining that, despite all of the craziness that's going on, I think that research into autism is something that the federal agencies are going to continue to prioritize. So thank you very much. [Applause]

Speaker 1: I have two questions. The first question is, is the de novo mutation is not the single cause? So there is an additional factor attached to that. I have my own theory for that. I have two kids. One of them in a spectrum. I always hypothesize that probably there is a double heat, second heat. Definitely, it's not vaccine, but there is something. What do you think could have caused that large-scale regression? Because it might not be just one gene. If there is a de novo mutation, then there must be many genes got dysregulated some way, and then there is a manifestation of ASD. What are your takes on that?

Jonathan Sebat: If you just focus on the one gene and just the de novo mutations in one gene, what you'll see is that only about 25%, everybody with those mutations will have symptoms. There'll be symptoms, but only 25% will meet full diagnostic criteria for autism. It's not sufficient. The gene by itself is not fully sufficient to meet all the diagnostic criteria for autism. It does a lot of things. The second hit that you're talking about, there are second, third, and 100 additional hits that are in a more subtle fashion shaping clinical outcome of these mutations. It's never just one thing. Even if you had a genetic therapy that would reverse the causal mutation, it would undo the effects of that one gene. It would not change your identity. It would not fully change everything about you, and all of those other aspects will still be present. A genetic therapy for that one gene would fix very specific neurodevelopmental effects of one gene. But like I said, autism is even if you have one of those mutations, that gene alone is not the entire story.

Speaker 1: Just a follow-up question to that. In a family, when you have like two kids, they're born within, let's say, two-year apart. One kid is extremely neurotypical, smart, and brilliant, but the other kid is ASD. Is there any clinical trial or research done on that? Because that can, per my case, I think all of them equal. There was no sudden environmental change, no a change, but that is not only two years. Nothing changed within two years. They went to the same pediatrician have the same lifestyle. Everything remains the same, all else equal. But one kid is on the extreme end of the spectrum, other kid is brilliant.

Jonathan Sebat: These are not identical twins, right?

Speaker 1: No.

Jonathan Sebat: There are many other things that are shaping the outcome. Even if there was one mutation that you knew about that they shared, there are a lot of other things shaping outcome.

Speaker 1: Thank you.

Speaker 2: I actually had a question if your group is exploring epigenetics with these genes.

Jonathan Sebat: Yes. There's a new technology on the block. Illumina, down the road, developed the technology that we're currently using for all of our sequencing. But there's a new kid on the block, Pacific Biosciences and Oxford Nanopore, and these are very different types of sequencing now. They sequence very long pieces of DNA in one single strand. You can actually see the methylation on the DNA as it comes out. It's really changed the way we interpret genomes in a big way. We can see mom's chromosome and dad's chromosome. We can separate them because these things are so long that we can phase them apart. We can read all of the methylation around the gene, and we can see

that when there's a mutation, does it change methylation patterns. Fragile X is a perfect example. Fragile X gene turns itself off when that Fragile X repeat gets so big that it's heavily methylated, and it silences the gene. That's an epigenetic effect. Fragile X is a perfect example of an epigenetic mechanism that's caused by a genetic change, and that happens all the time. We can also see when a mutation happens on an X chromosome and they have X-linked dominant, like Rett syndrome, where it's mostly girls who are affected by the X-linked mutation. We can actually see from the methylation patterns that mutated X is actually heavily skewed and that the active. You know how women have X inactivation? Every woman has a different X chromosome being expressed in every cell, but only one. That pattern of inactivation gets completely skewed when you have this mutation on the X, and you can see it clear as day in this kid [inaudible 00:35:55] because you have all that methylation. We get so much more information now from these new sequencing technologies that you get that epigenetic data.

Speaker 2: On the environmental aspect, is it true that the Amish community have really low autism diagnoses, and are there other communities where you find that?

Jonathan Sebat: There's actually many, many genetic disorders that were discovered in the Amish community. From the genetic standpoint, the Amish community has been a gold mine for finding genetic causes of disease because they have these big pedigrees, and no, it's not true that there's no autism in the Amish community. Again, we hear about how the increase in prevalence is really driven by diagnostic drift. Now, that diagnostic drift is not just increased awareness. This is just systematic differences in how health care is administered in one group versus the next, and it creates geographic differences in autism rates. California has a much higher rate of autism than Maryland. There are all sorts of reasons why that is. It has to do with the services, and of course, it wouldn't surprise anyone that the Amish community would access services in a different way.

Speaker 3: When you talk about mutations, older fathers and that and de novo, that's not as simple as redheads jump a generation or anything. We're talking about de novo, meaning the parent while the parents alive, not the grandparent or something from the past.

Jonathan Sebat: The mutation itself happens. In every sperm, there's 100 new mutations. It just happens. Every once in a while, it'll disrupt a gene. This is very normal everyday part of genetics that it's happening. It can skip a generation in certain ways. For example, it can occur in the developing embryo, so it's only present in maybe 10% of cells. It's actually present in mom and dad, but only in 10% of dad. It doesn't produce much of a phenotype, and then it gets transmitted, and it's present in 100% of cells after it gets transmitted. That's a way it could skip a generation. That's an example. But the more typical way that it skips a generation is that it's just not fully penetrant. That's the more typical way, is that the variant itself needs the second hit we were talking about. That the second, third, and 10th, and 100th hits really do shape the clinical outcome in a meaningful way. In the very first individual that had the de novo mutation, they were blessed. They were dealt a good hand in terms of all the rest of the genome. But then just by total chance, the random segregation of all of mom's genes and all of dad's genes can happen in myriad different ways, and so just a random chance can produce a higher genetic load in a child, combined with the mutation, and then suddenly you have a more severe clinical presentation. That's probably the more typical way that it would skip a generation.

Speaker 3: Do you have a sense of which chemicals or other thing might? There's so many things that could be influencing this. That's a whole thing we discuss.

Jonathan Sebat: Well, we recently did a study of smoking. We actually got scooped, though, because my colleague Hilary Martin in the UK published it before we did. But the smoking has the tiniest and tiniest of effects on mutations in both mom and dad. We can see it. It requires you to look really carefully. But smoking has a very tiny effect, and you can see that. But you can also see differences

between populations that are probably not genetic. They're probably environmental differences in the rates of mutation as well. African American populations have a slightly higher mutation rate than the European populations, and that's probably environmental. That's probably other exposures in the population. We've heard of air pollution as a possible risk factor, and that's one of the things we're going to study in our new grant. We are going to tease apart whether or not mutation could be part of it. Then, again, we're going to tease apart all of the rest of the correlation and causation. Thank you very much. [Applause] [Music]