ISAAC SAMUEL KOHANE: So what we'll talk about today is a very practical genomic medicine, by which I mean not only stuff that we think is going to be imminent in two years but what it means today to do genomic medicine, nothing future, just today. And if you think that I have stepped across a line into the future, call me on it. So let's talk about different diffusions of knowledge. The genome draft, as you know, was much heralded with Clinton-- was Clinton still president in 2001? No. So before-- when was the draft? The first--

AUDIENCE: [INAUDIBLE].

ISAAC SAMUEL KOHANE: When was Venter and Collins-- and wasn't Clinton-- yeah, it was Clinton.

AUDIENCE: It was 2000.

ISAAC SAMUEL KOHANE: It was 2000. That's an error. So it was much heralded. And there was a lot of promise about this would solve problems in human disease and in medicine. And it made it sound-- the way it was described-- as fairly imminent. And it was going to transform medicine. And just as a measure of calibration, the development of penicillin by Chain and Florey in '41, was saving thousands of lives within months.

And so has genomic medicine been successful by that measure? No. Do we think it's going to impact medicine in the future? Well, sure. I hope I've convinced you in the course of the class that it's going to be important in the way we diagnose our patients, in the way we manage patients, even the way we treat our patients, but clearly not by this measure. And I think we have to appreciate that probably timescales on the order of 10 to 20 years.

And, in fact, one of my colleagues, one of my friends used to be a reporter for the *New York Times* when this was announced in 2000, asked me, so what's going to be the main impact of the human genome draft for the next 10 years? I said, a lot of bad news. By which I meant that we're probably going to be able to diagnose a lot of things that we had not been able to diagnose and do nothing about it. And that's probably why not. But there's another reason why not, which is, well, what other problems have there been in preventing the diffusion of genomic medicine? Any ideas?

AUDIENCE: Massive amounts of data?

ISAAC SAMUEL KOHANE: What?

AUDIENCE: Massive amounts of data.

ISAAC SAMUEL KOHANE: Massive amounts of data and--

AUDIENCE: Lack of trained personnel.

ISAAC SAMUEL KOHANE: --and lack of trained personnel. Basically, the medical system does not have any idea how to deal with this. And just to give you some calibration around that. I gave a lecture a year ago to American Professors of Medical Genetics, not genomics but medical genetics. And I was telling them about the whole genomic revolution. And I think it's an odd thing for me to talk to professors of medical genetics and the national association.

But they were polite and interested in my lecture. But when I asked them, is this what you see yourself doing for the next 20 years? Absolutely not. They wanted to restrict themselves to monogenic, strongly highly-penetrant diseases that were extremely rare. That's what they wanted to do. And I said, well, you share at least a substring with genomics. Don't you see it as a part of your field? No. And so the geneticists are not holding the torch.

Right now, the medical students are not being taught this in any detail whatsoever. And so there's going to be a problem. So who is going to practice genomic medicine? There's a variety of possibilities. Shown in light blue, for some reason, is the medical geneticist. The person who typically has been doing-- sorry about that-- who has typically-- that will be the last light blue thing, I hope-- who has been doing medical diagnosis and counseling.

But what I'm telling you is they do not see themselves as bringing into the for the entirety of the impact of genetics into the broad swath of diseases, whether they're primarily inherited or primarily modifying environmental influences. A logical place would be primary care. Because, after all-- as I hope I've convinced you-- that, indeed, part of what genomics gives you is the ability to prognosticate for the future. Part of preventive care should be genetic and genome-wide testing. So a pediatrician, internist, OBGYN would be a natural place for this to happen, and also for specific diseases.

We've seen a lot of application of microarrays to cancer, for instance, so oncologists would be the natural person to order these tests. And a gastroenterologist-- we looked at Crohn's disease. And it would not be illogical for gastroenterologist to screen for risk factors. So what's, in fact, the case? So there was a study done just looking at cancer susceptibility tests. So it's a substantial study looking at over 1,200 physicians, 820 of which were in primary care.

And in 12 months, approximately 30% ordered or referred genetic testing, looking for susceptibility, not looking for diagnosing a patient but for looking were they at risk. So that's kind of impressive. So a bit less [INAUDIBLE]. Only 7% of them directly did it, whether because they were uncomfortable or not knowledgeable enough to actually directly order it. But that's telling you that 30% of this random sample of physicians were actually ordering these susceptibility tests. What do you imagine are the factors affecting ordering? Let me give you some potential-- what were you saying?

AUDIENCE: Cost.

ISAAC SAMUEL KOHANE: Cost. Any other suggestions?

AUDIENCE: Whether you can interpret the test.

ISAAC SAMUEL KOHANE: Whether you can interpret the test. Any other suggestions? What?

AUDIENCE: [INAUDIBLE]

ISAAC SAMUEL KOHANE: Which physicians were more likely-- no, no. What I meant was which physicians were more likely to order tests. I apologize for-- let me rephrase the question. What was it about them? What property of the physicians made them much more likely to.

AUDIENCE: Probably research hospitals.

ISAAC SAMUEL KOHANE: Research hospitals.

AUDIENCE: And also probably in disease areas where there's a lot of--

ISAAC SAMUEL KOHANE: So places where they have a high burden of genetically-- disease. Well, here's the answer. The first and foremost was being in the Northeast. I don't know what it means. But that's, in fact-- it was a by far-- so there's plenty of wonderful tertiary care centers on the West Coast, I hear. But that was not a telling thing. Feeling confident that, as you mentioned, the presence of advertising materials and, most importantly, having the patient ask for it.

The point here is that the reason there was such a high percentage, 30%, is not because doctors have been trained to order those tests. It's because the patients are reading lay literature. And if they have breast cancer in the family or have ovarian cancer, or the have colon cancer, they're asking the doctors to test them. That's the real insight.

[SIDE CONVERSATION]

Go ahead.

AUDIENCE: When you say susceptibility test-- presumably some kind of multiplexed PCR looking at different [INAUDIBLE] or associated risks that actually [INAUDIBLE].

ISAAC SAMUEL KOHANE: It's a multiplex test not an array test.

AUDIENCE: Question about Northeast-- is it just simply medical professional density?

ISAAC SAMUEL KOHANE: No, this corrected for it.

AUDIENCE: [INAUDIBLE]

ISAAC SAMUEL KOHANE: Yeah.

AUDIENCE: The *New York Times* or [INAUDIBLE].

ISAAC SAMUEL KOHANE: It's something else. It's corrected. But it's something about the nature of the training or it may be patients in the Northeast. Let's put it this way. Maybe there's a lot of Ashkenazi, Jewish women worried about breast cancer knowing that they have this BRCA1, BRCA2 risk factor. The article did not actually elaborate on that. But that's my guess. I think the patients are the driving factor.

And for those of us who are in medicine, I think we have to recognize this. And it's actually a sad fact that it's the patients that are driving it. But it's a problem because if one of the things that you bring to bear, as a doctor, is an appreciation of prior probability and what tests you want to do in order to avoid false positives. Because, remember, a lot of these polymorphisms or mutations may not actually be the causative element. They may be in linkage disequilibrium, for instance.

And so it's not a 1 to 1. It's not if you have this, you're going to have cancer. It's a probabilistic measure. And so there is going to be a false positive rate with any of these measures. And if the doctor is really not knowledgeable about that interpretation, then they're going to find problems. Just as if doctors did the routine CT scans on everybody, you'd find on the order of, people my age, maybe 10% to 20% pituitary microadenomas, as we find when we do autopsies of car accidents in people my age.

But they have no clinical meaning that we can tell. But if you do routine testing, you'll find that. And this is going to be quite a huge problem of false positives if we continue to have patient-driven demand for testing, because it should be done knowledgeably.

AUDIENCE: Can that be met with if a doctor orders this or refers them to a specialists [INAUDIBLE]. When these tests come back, I'm assuming you don't-- this is probably wrong-- that they don't come back the way the normal blood test-- there's certain numbers and whatever the doctor's-- blood tests.

ISAAC SAMUEL KOHANE: The short answer is theoretically not.

AUDIENCE: [INAUDIBLE]

ISAAC SAMUEL KOHANE: No, there actually has to be person. We'll get to that. But even so, if you have an ascertainment bias because of who's doing it without actually letting the medical system try to define a appropriate measure of when you do a test, then you're going to run into those problems more than less.

AUDIENCE: Oh, so you're suggesting that maybe-- so patients are asking for a particular test because they're worried.

ISAAC SAMUEL KOHANE: They're worried.

AUDIENCE: So you're suggesting that maybe that's a bad thing not because the doctor should be the ones to say, hey, you might want to think about this. But you're saying that maybe they--

ISAAC SAMUEL KOHANE: It's a self-selected group. And it may be that the people who are truly at risk are not getting tested. Basically, the doctor is not a decision maker here. And the whole role of a doctor is to decide, if anything, when is a test going to be cost effective and sufficiently useful for the patient. And if they disintermediate themselves and allow the patient to do that, then there's a risk that there's a group of people who will not be screened and a group of people who have been over-screened and, therefore, have an unacceptably high false positive rate.

Because, basically, when people come up with calculations of sensitivity, specificity, they do it based on a certain population of patients, when a genetic counselor says, you have X, Y percent risk based on a general population. They don't based on the group of patients who are neurotic and worried, maybe appropriately, about their family history. That might be a different set of specificity and sensitivities. I'm not saying that patients are wrong to be worried. What I'm saying is the medical system is failing. They don't have educated doctors who can intermediate about what is the appropriate testing to be done.

I mean, if a patient comes to me as an endocrinologist-- says, I want growth hormone testing. I not to do growth hormone testing when everybody asks because they have a significant false positive rate. And they'd end up treating a whole bunch of people with growth hormone for no reason. And that's the whole point of having an expert as part of their loop. And what I'm saying is this is telling us that there's a demand. And the medical system is failing.

So here's the conventional view of genetic information management. It's worth going through it. You can think of disorders being single-gene or chromosomal, major gene or multifactorial, or these complex traits, complex multifactorial. And the conventional view of how this should be used is, in primary care, the point would be to recognize signs and symptoms, make referrals, support family long-term care. The primary care should appreciate the role of family history, arrange testing, referral to specialists, as needed, provide longitudinal care. And for complex traits-- use of genetic tests to guide prevention treatment.

The specialist would manage specific problems. They would diagnose and manage system-specific problems. And they'd use also genomic tests to guide, prevent, and treat. And the medical geneticist-- the role there is counseling, longitudinal care, advice and interpretation of test results, and a reservoir of knowledge of handling of complex cases. But I just told you that medical geneticists have largely not met this role. So is there a different role? What to do. Is there a medical genomicist? What do you think? Is that a reasonable specialty or is that the wrong answer.

AUDIENCE: Explain what a medical genomicist means.

ISAAC SAMUEL KOHANE: If you wanted a good preventive care and you wanted someone who would give you appropriate counseling about either lifestyle changes or appropriate drugs that you need to consider, or a specific diagnostic test that you have to do for all possible genetically influenced diseases, what kind of person would you what you want to go to? Any ideas?

AUDIENCE: Actually primary care is my first reaction.

ISAAC SAMUEL KOHANE: So your first reaction is primary care. Do primary care practitioners currently know about any of this stuff? No.

AUDIENCE: They [INAUDIBLE] with someone who would give them a very simple explanation. If someone has this test and [INAUDIBLE] what it means. And they're going to tell you what it means and then manage that through your whole--

ISAAC SAMUEL KOHANE: So let's do it slowly with feeling. Because you're now the czar of medicine. And you're trying to start a company in this area, in the United States, in 2004. Who's going to actually do this job?

AUDIENCE: Well, I think the idea of going to any one, single person, whether it's specialist, genomicist, geneticist, or primary care-- I think is the wrong approach. I don't think any one person-- just in their specific roles can address-- especially as you move down that leftmost column to the very complex disease. I think that's the--

ISAAC SAMUEL KOHANE: So what do you do? We know that there's a large number of people who are going to die of complications of type 2 diabetes. And it's exploding. We know there's a large cancer burden. We know there's a large cardiovascular disease burden. And we know that some component of all of these diseases is, in fact, down at the bottom. So what are we going do?

AUDIENCE: So I think rather than having a medical genomicist, it would be nice to have a type of physician who organized with different levels of care into a seamless approach to be able to [INAUDIBLE] approach, to address the [INAUDIBLE] you have. Maybe the genomicist would be appropriate for homing the genetic aspects [INAUDIBLE] collaboration. You have somebody who is seeing a patient, outside of their regular PCP, who can integrate the latest technologies along with the traditional therapies and can communicate the process--

ISAAC SAMUEL KOHANE: So basically someone who's a generalist in medicine but a specialist about inherited diseases, and complex diseases, and also a team communicator, coordinator.

AUDIENCE: Right. So for each patient, you've got one person who's not having to worry about the occasional sniffle here or there, like a PCP, but also is focused on a bigger picture than maybe the specialist or the geneticists.

ISAAC SAMUEL KOHANE: Go ahead.

AUDIENCE: I think it's unrealistic to have a team-- I wish I had a team of specialists around me all the time to do this stuff for me. I think the reality is you're going to have, at most, one person who knows anything about your health, any ongoing basis. And that person should be the counselor that let you know things about lifestyle, things about preventive care.

And they should have genomic information to help give you advice about your particular lifestyle decisions you make. And that should be the primary care physician. So I think that it's going to be really hard to have a lot of people around, these specialists like genomicists giving you this preventive care type information. Because they don't that well.

ISAAC SAMUEL KOHANE: So you say the primary care practitioner. Cecily.

AUDIENCE: I mean, to be honest, more and more genomic tests come online. Like, look at cancer microarrays. You're just going to need people to know about the techniques and know about the methods. And you can help the physicians interpret it. I mean, there's going to be a role for them. We're seeing already there's going to be an explosion of SNPs, and pharmacogenomics, and all this stuff. And you're going to need to have people who know the techniques who can help the primary care--

ISAAC SAMUEL KOHANE: Absolutely true of what you both said. On one hand, it's unlikely we'll have swarms of people worrying about us. And yet, there is going to be this explosion. And no one person-- I mean, it's already pretty hard to be a good primary care practitioner with knowledge circa 1990. How are they going to do that? Any ideas? Operationalize what you were suggesting.

AUDIENCE: You might have, in a hospital, a medical genomicist that these primary care physicians can call up and refer a patient to the help interpret things.

ISAAC SAMUEL KOHANE: But what if it's primary care? I mean, we're all going to, unfortunately-- unless one of is a major messianic figure-- going to die from one disease or other. So we're at risk. And, hopefully, it will not be a bus hitting us. So it's going to be something that's going to come-- interaction between the environment and our genes.

So that's for all of us. So, basically, we need that information, unless we don't want the information, for every one of us. It's not going to be, hey, I have an interesting problem. It's part of routine care, I think. Everybody needs to be--

AUDIENCE: There could be a department of genomics in any--

ISAAC SAMUEL KOHANE: That's the academic answer. I'm talking about the process in the field. Who's going to be the guy or girl who's going to be providing that knowledge, either as a primary care practitioner or to the primary care practitioner? So here's some ideas I had. Your ideas are-- by the way-- are as good as mine. And I want to tell you, this is actually the central-- I mean, this course is about genomic medicine. And I think this is a central conundrum.

There is no plan right now how to bring all of this into the field. And I can tell you, in pediatric endocrinology, we're not even ordering the autosomal-dominant, well-understood, highly-penetrant single genes that we know about because of the educational problems, let alone all these other complex diseases. So any idea that you have could be of great interest to our system or to a company, if you could actually figure out how to do this right.

AUDIENCE: Some of it is going to either market-driven--

ISAAC SAMUEL KOHANE: It's definitely going to be market-driven.

AUDIENCE: --I mean, right now, [INAUDIBLE] done a lot of research in but there's a really good test and really good results come online and generate high-profile papers and get into some of the patients are going to start demanding these things.

ISAAC SAMUEL KOHANE: But that's a problem. For instance, a prostate-specific antigen done wrongly gets a bunch of poor guys get their prostates removed for nothing.

AUDIENCE: [INAUDIBLE] error too, 7% false--

ISAAC SAMUEL KOHANE: False positives.

AUDIENCE: --false positives.

ISAAC SAMUEL KOHANE: Yeah, and so that's the problem. And, as I explained to you in my first lecture, there's 7,000 articles just on appropriate use of the prostate-specific antigen. We have now 30,000 genes to worry about. And what are the right cutoffs. And let's be optimistic and say we can actually get the optimal answers for these. No one individual I can think can actually solve it.

So you're absolutely right. It will be market-driven. That's certainly true. But the question is, how will the market solve that problem? And I'm claiming there are companies out there that will solve that. And either it'll be inspired by ideas similar to ones we're going to articulate today or by other ideas. So all I've been calling for is thought about this. So one was internet-enabled triage specialists.

Basically, you feed your genome to, essentially, a service on the internet. And they take whatever the primary care physician says and they basically-- in India or somewhere else-- provide you with your risk profile and the next steps for you to take. That's one possibility. So you check each patient in door for inheritable genetic or epigenetic phenomenon. A parallel system-- that's, essentially, a fly on the wall to a mental interview, provide decision supports.

And this could be a person or, more likely, a computer program. But it's going to don't have to be a damn good computer program to be able to do that. But I was involved in knowledge representation and knowledge-based systems in the 80s when I did my thesis. But there really wasn't any good motivation for it. Because, in the end, doctors know how to diagnose acid base problems.

But doctors will never be able to do this. So this is a true motivation, I believe, for automated decision support. The alternative is just to redo the curriculum in a major way, and even so, to teach students how to use electronic resources in near real time. Because no one will be able to keep this in their head.

AUDIENCE: For the last one, I think it would require such simplification of outputs in terms feeding a genome in and spitting out numbers, the way that [INAUDIBLE] and all these other things, because that's how-- right? I mean--

ISAAC SAMUEL KOHANE: But it would not be a number. When I was talking about when you're feeding the genome [INAUDIBLE]. The answer would not be these are the levels of your various things. It would be you have a 30% increased risk for prostate cancer. The following test is the right thing to do, that kind of thing.

Because, again, no primary care physician can know it. As I said, almost none of them know fully all the literature and prostate-specific antigen screening for prostate cancer and let alone for 30,000 tests. So I think it would have to be a distilled, utility-based, sensitivity, specificity-based--

AUDIENCE: That's what I mean. They get a [INAUDIBLE] from the lab. They get it from the--

ISAAC SAMUEL KOHANE: --oh, it would be simplified in that sense. Absolutely. I think that is the only way. And endocrinologists have made a big career out of just interpreting three numbers-- TSH, TBGI, and T4 in the thyroid test. Because guess what? Most medical students can never figure out which way means hypothyrodism, hyperthyroidism, or changes in the amount of binding protein.

AUDIENCE: Doesn't that then relegate the medical geneticists to just the category of specialist once again? the lack of a specific disease--

ISAAC SAMUEL KOHANE: I'm claiming that the system cannot have a expert medical genomicist. Because a lot of what we were talking about is primary care, is prevention. This has to be in every primary care situation. So it's either outsourced, essentially, to the internet, to some other group of people, have a low-cost parallel assistant, whether it's a human or, more likely, a computer program. And the third is redoing education.

To your scenario, which, I think, is one of the good ones, which is to have a very simplified set of recommendations come to the primary care physician. It's the primary care physician, still, who just has to learn how to look intelligently at that report, that set of recommendations. I don't see them having to learn the 30,000 genes and all the epigenetic and genetic effects. That does not seem reasonable.

But this is the fundamental problem that we have in genomic medicine. And that's going to be, by far, the rate-limiting step for any penetration to managing human disease than any other aspect of the genomic enterprise. We're going to discover lots of interesting things. We're going to find good drugs. We're going to make interesting diagnoses. But translating it into cares will be the real challenge. So how are we going to pay for those roles? Extremely unclear. Right now, you can't get a lot of genetic tests paid for.

AUDIENCE: [INAUDIBLE] because it hasn't been developed enough in terms of [INAUDIBLE]

ISAAC SAMUEL KOHANE: Right.

AUDIENCE: [INAUDIBLE] combined [INAUDIBLE] result.

ISAAC SAMUEL KOHANE: With the result. And, finally, because it's very expensive. And if it became free, everybody would use it. I mean, right now, as I told you the first lecture, in a high-throughput lab, it costs $0.10 to do a genotype. Do you know how much the system is built for, for that test? So I was talking about, at first, $0.10 in order to tell you, wow, the genomic future is now. We can do very cheap screening.

But how much is our insurance going to be billed for these tests, for that single genotype?

AUDIENCE: Over $1,000.

ISAAC SAMUEL KOHANE: Over $1,000.

AUDIENCE: [INAUDIBLE]

ISAAC SAMUEL KOHANE: And that's not the real cost. That's a huge profit margin.

AUDIENCE: There's been a similar debate going on with these full-body MRI scans and things, right?

ISAAC SAMUEL KOHANE: Sure.

AUDIENCE: And so, I mean, where are the voices in the field for this and for those saying, preventatively, if we do a whole battery of non-invasive, one-day tests- submit a sample that you can do with genomic testing. You sit in the MRI and do all the stuff. And then, hey, we found this particular disease. And we just saved 60, 70 maybe $100,000 worth of treatment.

ISAAC SAMUEL KOHANE: Well, first of all, that's not the way the system sees it. The system sees that you spent $4,000 bucks. And my insurance company is going to reward me for what I do the next two or three years. If you drop dead of cancer 10 years from now, that's going to be the balance on the insurance company 10 years from now. There's not a lot of payback to me.

But more importantly, speaking as a clinician, I think these total body MRIs are terrible. Because you're finding a lot of incidental findings. In fact, the medical establishment is against those, I think, for good reason. Because, again, the probabilities that we have around the meaning of bumps on the MRI is around people presenting with certain symptoms. Because these studies are aimed around that.

And if you just have Joe Blow off the street who just has a birthday present, a full-body MRI scan-- which, by the way, haven't you heard the advertisements for it? Yeah, it's your 40th birthday. Don't you want to do this? Or your dad's birthday-- and the advertisement is Joe Blow, the patient, reports, oh, I'm so reassured.

On my 40th birthday, seemed nothing's wrong with me. What they don't report on is all the people who got bumps in their head and then had to go through craniotomy, God knows what to investigate these things. So I promise you that we will stick to the pragmatics. So this is why there's not penetration today. But today, how do you order a genetic test for a clinical problem? What do you do?

AUDIENCE: How do you order it?

ISAAC SAMUEL KOHANE: Well, yeah. What do you do?

AUDIENCE: You go to geneclinics.org and you check if it has it and whether it's a research lab or clean up--

ISAAC SAMUEL KOHANE: Did everybody hear that? That's very, very important. Gene clinics and genetests.org. It's one of the things better stick to you after this class. Because that there are thousands of clinics and labs across the United States that have one or two tests. There's no single, giant aggregator of genetic tests. And there's this one website, maintained by my colleague, Peter Tarczy, at the University of Washington, that has all that. So that's exactly right.

AUDIENCE: I mean, just a question of the market stuff. Do you think that's going to eventually roll into the [INAUDIBLE]

ISAAC SAMUEL KOHANE: 100%. So that's an excellent question. OK, let me just get to that question in two minutes. Sorry, it's an old slide. It's a year-old slide. And they had-- on gene clinics, gene tests-- 189 gene reviews, which are very nice monographs around a specific disease involving 1,000 different clinics and 500 laboratories covering 948 diseases.

And the short answer is, yes. These are going to be rolled up into requests and the various laboratory corporations. The real question is-- ultimately, we know that what it's going to look like. It's going to be a highly-roboticized sequencing genotyping operation in these labs. But in the interim, it may still be that it's rolled up. But the actual sequencing and procedures might still be at these various places.

And these companies might just contract to these various sources. Right now, for instance, if you want to do Duchenne testing, your sample goes to a place in Utah. Now why is that? Why not just roll into one place? Because the guys who are resequencing the Duchenne gene for Duchenne muscular dystrophy know which primers aka which sequence works the best to amplify different segments of the gene to get reliable results. And that know-how ultimately can be definitely rolled up.

But in the interim, on the five-year timescale, may or may not be just left locally. And so what these requests will be are more aggregators on front ends, conduits or channels-- as the marketers like to say-- to these myriad labs. Ultimately, I think everybody believes they'll be one highly-roboticized facility, each one of these facilities. Because after all, a gene is a gene is a gene.

But because of the things like knowledge of what primers work, knowledge of which mutations are common and what they mean-- all that knowledge management is still distributed. And rolling it up is going to be, I think, the brake on that full roll-up. So for those of you who don't know the site. So if you want to test for a particular problem, you can look at this thing and say, I want to look at obesity. And it'll tell you which laboratory will do which testing it for.

In fact, probably the only obesity-related gene worth testing for right now is MC4R. It's the most common genetically associated cause of obesity. MC4R is melanocorticotropin receptor, fourth type. And some high percentage, on the order of 2% of individuals with-- according to some studies-- with morbid obesity have mutations here. Unfortunately, for the rest of us, like me, we're just fat because we eat wrong. So we're talking about-- go ahead.

AUDIENCE: Geneclinics.org?

ISAAC SAMUEL KOHANE: Yeah. But is this indexing, that gene clinics and gene tests are generally sufficient. So if you look at adrenal hyperplasia, which is a disease of children and adulthood. It's responsible for ambiguous genitalia in infants, hirsutism, and infertility in females, and precocious puberty for males and females. So if you look for adrenal hyperplasia, you see the following. You get 21 hydroxylase deficiency.

But what's the right thing to actually order? Well, if you actually knew something about the pathway, the steroidogenesis pathway, you'd know that it starts from cholesterol. The cholesterol that we all claim to hate actually is the backbone for all the steroid hormone molecules. Cholesterol will make all the salt-retaining hormones like aldosterone, the glucose-stimulating hormones like cortisol, and the sex hormones like testosterone and estradiol.

And an appreciation of what the pathways are is going to allow you to actually do focus testing so that, for instance, 21 hydroxylase deficiency is, in fact, the most common. But there are other deficiencies that will happened with some regularity. 5% of congenital adrenal hyperplasia is 11 hydroxylase deficiency and not 21 hydroxylase deficiency. And the bars here just show the block in the pathway that will be caused by the deficiency of that gene.

And so the point is, you still need a lot of knowledge around that specific disease and the pathways rather than just going up to a database that says obesity-- this gene. You still have to have some knowledge about what you're looking for. So what material do we want to test? Let's take four diseases-- cystic fibrosis, a disease where you get plugging of mucosal ducts. And it's a chloride transporter problem. And people die young.

McCune Albright disease-- a disease of the G protein complex where, essentially, you have an activating mutation in one of the subunits so that all the G-coupled processes are hyperactive, so that you have kids with precocious puberty, cortical adenomas of the adrenal, and precocious puberty.

AUDIENCE: [INAUDIBLE]

ISAAC SAMUEL KOHANE: Well, McCune Albright syndrome, the classical description is cafe au lait spots, which are these-- basically, looks like someone spilled coffee on your skin. Fibrous dysplasia of the bone-- so your bones are screwed around because they have these sort of ropey things going through them, and precocious puberty.

AUDIENCE: [INAUDIBLE]

ISAAC SAMUEL KOHANE: It's distantly. As we learn more about it, multiple systems, like thyroid and growth hormone, can also be hyperstimulated. So, basically, it's a mosaic state where these mutations can actually be spread incompletely throughout the body. And so, for instance, the melanocytes are stimulated in just patches. And if you're unlucky enough to have adrenal involved-- adrenal adenoma. Please.

AUDIENCE: Can I get a scenario for--

ISAAC SAMUEL KOHANE: So a patient comes to you with McCune Albright syndrome. And you suspect it because Zach told me that they have these cafe au lait spots. Boy, this person has a two big ones. And this person is having a precocious puberty and is complaining of bone pain. What do I send to test? That's the question.

AUDIENCE: But whereas with the question of cystic fibrosis, they would know that [INAUDIBLE]

ISAAC SAMUEL KOHANE: Well, you suspect it. You did a sweat test, which has a certain specificity and sensitivity. or maybe it's a parent who already had a kid who died from cystic fibrosis. And they just have a newborn. What are you going to test? You don't know the N thing yet. So let's start one at a time. Cystic fibrosis.

AUDIENCE: [INAUDIBLE]

ISAAC SAMUEL KOHANE: A testing gene. But I'm a I'm a stupid doctor-- indeed I am. What does it mean to test genes? What thing do I stick into a tube?

AUDIENCE: Oh, just do a mucosal swab.

ISAAC SAMUEL KOHANE: So [INAUDIBLE] I can do a mucosal swab?

AUDIENCE: [INAUDIBLE]

ISAAC SAMUEL KOHANE: The short answer is yes.

AUDIENCE: [INAUDIBLE]

ISAAC SAMUEL KOHANE: What?

AUDIENCE: [INAUDIBLE]

ISAAC SAMUEL KOHANE: No. McCune Albright. Maybe the cells here are not involved. Maybe it's not--

AUDIENCE: You look for the [INAUDIBLE].

ISAAC SAMUEL KOHANE: Maybe you get a skin biopsy of the cafe-au-lait spot. 21 hydroxylase is efficient germline. You can do the blood. Clots like cancer, blood is not biopsy. You need a hunk of that cancer. Or depends, is a highly inherited cancer or do you believe it's a somatic mutation in the cancer? All the point I'm making here is it's not obvious what you put into the tube.

And as dumb as it sounds, when you're a tired resident on the ward and someone says, do the geniculate test. You don't know what the hell to do. It's that simple. But this is what it comes down to in genomic medicine. These are actually answerable questions, but you need some reading. Where are the mutations? And what do they mean when they are in certain tissues?

AUDIENCE: So diseases [INAUDIBLE]

ISAAC SAMUEL KOHANE: For a germline disease-- any tissue. It doesn't matter. So you want one that is the least invasive-- blood, swab, even spit sometimes.

AUDIENCE: Right. For some other complexes, [INAUDIBLE] people suspect you have susceptibility, which then triggers the disease.

ISAAC SAMUEL KOHANE: It's going to be certainly not true for many diseases. But, for instance, when Scott Weiss talks about asthma, he's thinking of a risk factor, even if it's a small risk factor and it'll increase your risk of asthma by 1.2, he's still thinking of this as a germline polymorphism. Now it may be that people that asthma-- I'm just making up something that's completely off the beaten path-- is, in fact, a somatic mutation. I doubt it. But all these genotyping studies, haplotype studies are done off of blood. So they're assuming these are germline diseases.

AUDIENCE: So for something that's not germline, something like maybe you've got a disease because you're exposed to some sort of environmental toxin or something--

ISAAC SAMUEL KOHANE: Or you have a somatic mutation. Yeah.

AUDIENCE: --yeah. Would it be a good standard protocol then to take-- let's say for cancer, part of the tumor and part of just any other tissue to compare--

ISAAC SAMUEL KOHANE: Or blood. Sure.

AUDIENCE: --or something like that just be able to--

ISAAC SAMUEL KOHANE: The short answer is cost. But, yes, I mean, the answer is--

AUDIENCE: --wasn't it Weiss who said that in the next five years this cost thing is not going to be an issue--

ISAAC SAMUEL KOHANE: Well, it's not going to be an issue for him doing research. What I just told you is $0.10 a genotype today for Scott Weiss. For you, my friend, $1,000 in the clinic. And by the way, that's a huge commercial opportunity obviously. You're absolute right. That's going to drop. It's like a drop to $0.10. It's going to drop to $10 or $100. And the market leaders in that, who can figure out how to make the bucks on this, at that level, are going to be extremely rich.

OK, so if you ordered a test, you going to need to get advice on that genetic test. Where are you going to get that? Found either in your office-- let me call you off the bat-- extremely unlikely. There's no one in my office who's going to give you genetic counseling or other licensed counselors-- maybe it's part of your institution. I'm talking about today. We're not talking about the medical genomics.

Today, what are you going to do? Or licensed counselors at Brigham, or you outsource it-- and here, it says, there's a pre-test phone consultation provided patients and so on. And that might be part of the business model, that they understand that a lot of institutions don't have that facility. So they can outsource it. So how do you send a store for something.

Again, these are stupid things that most residents don't know. 310 milliliters of whole blood, if it's a germline in a purple top tube, also acceptable green, which is sodium heparin, or light blue-- sodium citrate tubes. It comes down to that. What tube you put it in? If you put it in the wrong tube, you may not have an adequate extraction. And it's totally different, of course, for RNA. If you're interested in RNA, you better flash freeze it, is a short answer, as soon as you can. So what kind of DNA--

AUDIENCE: [INAUDIBLE]

ISAAC SAMUEL KOHANE: Because, basically, the amount the stability and extractability is going to depend on what is inside the tube. And the color of the tube is a code of what's inside the tube. So these tubes are not all featureless glass. They have a little bit of chemical in them. So what kind of DNA testing should be done? So let me ask you guys.

We've just got together in a happy company called 512 Genomic Testing. We've got our stock options. And what are we going to do? Are we going to sequence these genes? When someone sends us a gene to 512 Genomics, are we going to sequence it or are we going to genotype it for all the known mutations? What are we going to do?

AUDIENCE: [INAUDIBLE] mutation different levels [INAUDIBLE] and more complicated.

ISAAC SAMUEL KOHANE: So let me push you. So just arrived in our inbox is a blood sample in the right tube from Joe Schmo, but it's really her baby's blood. And she wants to know, does this baby have 21 hydroxylase deficiency? And they heard that 512 Genomics is a very smart company because they've been trained at Harvard Medical School. And, therefore, they trust us to do it. So you're going to do a panel of what? Genes? Let's simplify your life. Well, first of all, are we going to test all genes?

AUDIENCE: No, we'll start testing the genes that are known and that are frequent.

ISAAC SAMUEL KOHANE: OK, I want us to have a very nice corporate vacation in Bermuda. So all the money that we spend is going to take away from the bennies I can give you on our vacation. OK, we're going to do-- I'm going to tell you this-- five genes that are [INAUDIBLE]. We're doing all five genes?

AUDIENCE: No, then you look at the frequency of a particular mutation occurring.

ISAAC SAMUEL KOHANE: OK, so I'm going to tell you there's two genes that account for 99%. Good enough for us? OK, let's say yes. Let's say yes.

AUDIENCE: Why can't we just do one gene, if it comes back negative-- if we know two compromise 99%, [INAUDIBLE] do the one, comes back negative, we do it. But if it comes back positive, it's like, hey, we got this. [INAUDIBLE]

ISAAC SAMUEL KOHANE: We're going to go back to the patient? That doesn't happen. We're a lab. We're not the doctor. That does not happen. We cannot ask, unless you really want to change-- you live in the dream world.

AUDIENCE: No, I'm just saying-- no, you can't go back to the patients and get more blood. But--

ISAAC SAMUEL KOHANE: Just first sequence the more common gene. Is that we're saying?

AUDIENCE: Yeah, so if you got two genes, I guess--

ISAAC SAMUEL KOHANE: OK, so let me tell you. Listen to the facts. 21 hydroxylase accounts for 95%, 11 hydroxylase 4%. So we got 99% with those two genes. So what am I actually doing? I'm not the laboratory technician?

AUDIENCE: --yeah, do the 95% one.

ISAAC SAMUEL KOHANE: OK, and it's negative. What do I do now?

AUDIENCE: Then you do the next one. It comes back positive-- I don't know enough about the disease. So assuming that the positive--

ISAAC SAMUEL KOHANE: The positive is important, positive is very important.

AUDIENCE: --then you just running that actual test.

ISAAC SAMUEL KOHANE: OK, so you're doing a phase one. That's perfectly acceptable. It's probably means that, most of the time, we'll run both tests. Because most of the time, we'll probably get referrals of patients who don't have that disease. So it's a small but important incremental savings you just gave us.

You took Jose's advice and said let's only do one gene. And if that's negative, we'll do the second gene. But as I explained, most kids this was sent for will, in fact, not have the disease because it'll be something else. And we'll still have to do the second one. You had a question?

AUDIENCE: The question is this is a PCR test, right? Just to clarify.

ISAAC SAMUEL KOHANE: Well, we haven't gotten there yet.

AUDIENCE: OK. [INAUDIBLE]

ISAAC SAMUEL KOHANE: OK, so let's say that we take the modification of the Jose protocol. So what are we going to do? So we look at one gene and then two genes. So what actually are we going to measure? Are we going to measure the full sequence of these genes? Are we going to just look at the known published mutations or SNPs? What are we going to do?

AUDIENCE: [INAUDIBLE]

ISAAC SAMUEL KOHANE: Now what if I tell you that 2/3 of the mutations are extremely common and 1/3 of them are one of a kind.

AUDIENCE: I'm assuming we know something about the frequency of having the disease associated with the frequency of having that particular mutation. So if we know that a particular mutation affects the one person and has a very high incidence of progressive disease, then that's the [INAUDIBLE]

ISAAC SAMUEL KOHANE: OK. I'm Joe Schmo. My kid actually had one of those rare ones. I'm going to assume your pants off. I'm taking the vacation to Bermuda because you guys took-- there was a known rare mutation. Or even--

AUDIENCE: Right, again, this is back to what I said in the Jose modification. If you come back negative--

ISAAC SAMUEL KOHANE: I see.

AUDIENCE: --then you go back--

ISAAC SAMUEL KOHANE: All I'm saying is that refinement is still going to cause-- most of the time, you're going to end up doing all the mutations.

AUDIENCE: But the alternative is to just do all the mutations all the time.

ISAAC SAMUEL KOHANE: What I'm telling you--

AUDIENCE: [INAUDIBLE]

ISAAC SAMUEL KOHANE: What?

AUDIENCE: A third of them are kind of unique.

ISAAC SAMUEL KOHANE: Unique, are private mutations. So the short answer is, I believe the right answer is, you've got to sequence the whole thing. I mean--

AUDIENCE: There's no value in preemptive screening, say, for instance--

ISAAC SAMUEL KOHANE: It depends. If it's for a given patient around the specific problem, that's a very different situation from I'm a public health authority and I want to screen the population or I want to screen the population and just detect people at risk for this. But if you're a doctor with a specific question, I'm duty-bound to actually do maximum for you. And consequently-- period.

AUDIENCE: And it just seems-- maybe I misunderstood this-- but it seems kind of counter to what the current practices were, again, standard testing for things. when you have thrombosis, the first thing they do is they test me for the most common things-- [INAUDIBLE] deficiency, and all these different things. Because, well, if you've got that, then [INAUDIBLE] then we don't really need to continue. Because we already know you had a big problem. So that's, I guess, the big basis for my approach. Joe Schmo came and said, one of my kids has this thing. The fastest, cheapest thing to do would be to test them for the most common--

ISAAC SAMUEL KOHANE: So you see you can do that because the doctor can actually cut a protocol and actually talk this through with the patient-- we'll go over the most common ones. We're 512 Genomics. And 512 Genomics does not have a relationship with the patient. And the doctor doesn't understand genetics, so we already established. So again, I'm trying-- 2004, if there is a private mutation that the patient subsequently finds, we're toast.

AUDIENCE: Even if we don't [INAUDIBLE] back.

ISAAC SAMUEL KOHANE: When are we going to go back? No one's going to come back to us.

AUDIENCE: Take the genes that account for those qualifications and people.

ISAAC SAMUEL KOHANE: Well, basically, we're going to follow doctor's orders if they say we want if they say if they say I want to look at congenital adrenal hyperplasia. I think we have to look at every gene known-- we'll build up accordingly, of course. If they say, I want to look at 21 hydroxylase gene, we have to sequence the whole darn gene. Now if they specifically say, I want to look at just a mutation-- which I'll never tell us. I want a mutation in codon 32. Well, we'll do that genotyping.

But if the doctor is a standard doctor, 2004, and says, check 21 hydroxylase gene. We've got to sequence the whole thing. There is no choice. Otherwise, we're toast. Yes, so I think the only thing I missed from the slide for epigenetic things like methylation, we need to think about doing that. But for certain diseases involve imprinting, for instance, you want to look at methylation.

So pre-flight checklist. What is this going to take to get our clinic up and running? Now we're 5212 Clinic. We're not the company anymore. So would you or a staff member be able to be-- so I took this from some very good website. I wish I could tell you which one it was.

But one of the things that you need to do before going, would you or a staff member be able to be an advocacy resource? Is a patient-- and when appropriate-- a family prepared for either a positive or negative test result? Does the patient understand the medical, psychological, and social ramifications of the test? Do you have a referral less than appropriate specialist and/or genetic counseling services to resolve any issues that cannot be handled in the office?

And these are all things that we have to do as a clinic before we can even start ordering these tests. And the workflow is as follows. For a positive test, these are all the things we have to do. Now we're a primary care practice. And the interpretation of a positive result is different if it's a diagnostic test, a predictive test, a carrier test, prenatal testing, or newborn screening.

If it's a positive test and it's clinical diagnosis confirmed, it's predictive testing. It tells you have increased risk. If it's carrier testing, it tells you that you're a carrier. It's prenatal testing, it says that the fetus has a specific condition. And if it's newborn screening, it tells you the newborn has a disease. And the follow-up includes all the things you could imagine. For negative tests, if it's a diagnostic test, the symptoms are unexplained.

For predictive testing, the likelihood of showing symptoms is decreased. If it's carrier, it's highly likely that you're not a carrier. If it's prenatal testing, if the fetus was symptomatic, then it's unexplained. If it's not symptomatic, the chance for the condition is small. And newborn screening-- the newborn is not expected to have the condition. There's a whole bunch of follow up that does or does not develop from these different levels of use of the same darn test across these different clinical states.

And thinking about 512 Genomics, our old company, if we didn't know which one of the situations are involved, we'd have to practice a very defensive corporate policy to make sure we'd be in the maximal informed state for all conditions. So can I send a sample for microarray testing? We've heard so much about microarrays in this class. In the USA, the answer is yes for a research protocol.

This summer, you're going to over at Harvard Medical School genotyping with a resequencing array for cardic myopathies and for hearing deficiencies. These are custom chips from Affymetrix. And certainly for research protocols, we're using them for expression as well. Can you do this for commercial testing? The answer is no.

Roche Diagnostics, three months ago, tried to get approved through the expedited review of the FDA, the P450 chip, which has all the genes that are involved for metabolizing toxins and drugs. And therefore, it's a very good pharmacogenomic screening. And they were blocked dead in their tracks by the FDA. Unless they can appeal it correctly, they will have to go through full bore extensive FDA approval. And So why is it the case?

I can give you a lot of different reasons. But the short answer is the FDA does not understand this technology and does not know what to do with it. I was part of a panel with the FDA-- [INAUDIBLE] talking about it-- they have just agreed, right now, with big pharma, just on what is a data structure with which they can transfer results, let alone interpretation or analysis of the results.

AUDIENCE: So they're restricting things just because they don't understand it, even though it's not unleashing a new drug on the population that they don't understand the side effects. This actually doesn't do anything other than maybe give patients something to worry about--

ISAAC SAMUEL KOHANE: Let me make it very concrete. It gives you a result that the patient then has ovaries and breasts removed for.

AUDIENCE: I guess when you put in that--

ISAAC SAMUEL KOHANE: Yeah, I mean, that's what-- what?

AUDIENCE: --it's a classic example of what I've heard from bilateral-- [INAUDIBLE] for no real reason.

ISAAC SAMUEL KOHANE: --for no reason. But that's going to happen. Many things are going to happen like that. I mean, just going to the doctor-- I try to stay away from the doctor as much as I can, which is probably a bad idea. Because you probably should get routine care. But if every time you expose yourself to even an investigative procedure for the wrong reason, they're likely to find-- again, to the false positive reason-- things that are incidental but are going to create huge costs, and worry, and morbidity.

Diagnostic tests themselves can kill you. So let's say you are told you have a risk for this. And then you get a colonoscopy. A tiny fraction, probably one in 200,000 people, suffer bad outcomes from that procedure.

AUDIENCE: [INAUDIBLE]

ISAAC SAMUEL KOHANE: Yeah, those are pain in the you know what. So this is a problem. So the short answer is-- microarray testing, no. But there's nothing wholly about the FDA. In the Netherlands-- I don't know-- again, this is what I had heard last year around this time. And I don't know if it's happened. But they were claiming they were going into routine use of microarray for breast biopsy screening and evaluation. I have to do the research to figure out whether it's happened or not.

And there's, of course, the issue of consent. And the short answer-- as we'll get to shortly-- is that you have to do a fairly extensive consent process. However, if the patient is symptomatic, if they have a cancer, or they're having a heart attack, or they have dementia-- they're actually symptomatic, it's a much more simple consent process. All the consent issues that you've heard about in public press are about pre-symptomatic testing.

AUDIENCE: So why don't they just approve that the pre-symptomatic testing with the caveat or the [INAUDIBLE] rule that we're just going to follow and observe a patient? Let's say that we usually go in and get a full-body MRI scan [INAUDIBLE] I think you shared a story with someone about that. Found it. And try to do nothing [INAUDIBLE] complications. So rather than go in and cut someone's head open, or a colonoscopy and everything else, just observe the patient-- six months, a year, two years, three years.

ISAAC SAMUEL KOHANE: So let me tell you something about medicine. That's impossible. If you find something that looks bad on an MRI-- because even if you believe that it's only one chance in 100 that is truly a bad thing and something bad happens to that patient, not only are you toast with the legal system but you'll feel terrible.

I mean, it could make you very, very fidgety to have a patient with a finding that you were taught in medical school is badness. And we're told in 2004, medical education in 2004. We're taught that some bumps on MRIs mean bad things. So if you start changing your priors by screening everybody, then bumps do mean different things. But that's not the way we've been educated. So we kind of get itchy as hell.

AUDIENCE: So with the advent of new detection technologies for particular things that you context intelligence specific for this sort of problem [INAUDIBLE]

ISAAC SAMUEL KOHANE: Yes, except it never really does because-- well, that's not true. For certain things, we're going to get specific enough markers. I can imagine that a bump-- may be wrong-- imagine a bump plus a functional assay, like with a PET scan, will show that this is a highly rapidly dividing thingamajig and, therefore, it's a problem. Or it's totally metabolically quiescent and we can just observe it. So that's possible. Yes.

AUDIENCE: I just listening to [INAUDIBLE] talking about his idea of why don't we treat cancers before they actually become--

ISAAC SAMUEL KOHANE: He's absolutely right.

AUDIENCE: --and that, to me, falls right in line with this just what's the harm in doing this analysis.

ISAAC SAMUEL KOHANE: Well, I have the greatest respect for Judah. And I'm sure he'd would agree with what I'm about to say. It all comes down to what is the treatment and what threshold do you pick. So if you're looking at a certain angiogenic signature, whether it's in a polymorphism or something that you're measuring, what's your false positive rate and how toxic is your treatment?

If you're treatment's totally benign, of course, treat everybody and cheap and treat everybody. But that's almost never true. It's never completely benign and completely cheap. So you end up having to make decisions. So now for presymptomatic asymptomatic testing, consent is actually a very, very complex and drawn out process. So here's what is involved.

Again, this is 2004. It may change to be more rigorous or less rigorous, depending on whether or not the Genetic Privacy Act, which is now somewhere in Congress, will be enacted or not. So the current state-of-the-art is, the major medical facts including the diagnosis, the prognosis, and the way the treatment of disorder tested has to be explained to the patients. The genetic facts involving-- including risks for other family members has to be explained.

The chance that the test will give a correct prediction as well indeterminate or unexpected findings has explained to them. The risk of receiving an unfavorable test result and the possible consequences for yourself and your family-- in the case of prenatal diagnosis, this may include the risk of facing a decision about abortion. And you have to tell them in a way that they really recognize, they understand. Some people are videotaping these concerns now, well, defensively and also to make sure that the practitioners are doing the right thing.

Also, it should be made clear that can refuse the test and informed of potential benefits and disadvantages, including unsettled questions of privacy protection dealing with insurances, banks, or employers. You might be able to say today the insurance company is not going to do anything with it. But that test is now forever in your medical record and maybe not true five years from now. It might be a different insurance company or the same insurance company will feel differently.

Your care will not be jeopardized whatever decision you and your family make. Possible use of your tissue sample after testing-- they have to understand that. Whether it's destroyed or kept from an analysis and whether it can be kept for DNA banking. All of these things have to be discussed with the patient. This is not a five-minute discussion. And remember, again, back to our corporate clinic, 512 Clinics, our standard of visit for a patient is on the order of 10 to 15 minutes. And I'm telling you that this concept process requires an hour to do it adequately.

Having said this, I want to tell you, we're actually performing routine comprehensive genetic testing on our entire population. Did you know that?

AUDIENCE: [INAUDIBLE]

ISAAC SAMUEL KOHANE: You bet. So we're doing we're testing all kids in the United States for a genetic disease like phenylketonuria. We're now looking for all the common polymorphisms of CFTR, the cystic fibrosis responsible gene. And if you look what the Massachusetts State Laboratory is doing right now in Worcester with University of Massachusetts, I said, they're Jamaica Plain but they're affiliated with the University of Massachusetts. They're actually looking at maybe 10, 20 diseases that they're going to do risk factor screening for.

Now unlike our company, 512 Genomics, they really have to come to a couple of decisions about when to do this. How is it determined what is routinely screened? Three components-- public health assessment, evaluation of tests and interventions, positive and screening implementation. The public assessment is fairly straightforward. Disease or conditions should be an important public health burden not a rare bird.

And what does that mean? That means, typically, they like to see it above one in 20,000. They won't admit to that, but it's around there. Like thyroid disease, which was one of the first things to be screened, is about one in 5,000. Congenital adrenal hyperplasia, which they do screen for now, for 21 hydroxylase is about one in 20,000. The prevalence of the genetic trait has been known.

The natural history of the condition for susceptibility to link disease to overt disease should be adequately understood. And, basically, the safety and efficacy of the test-- blah, blah, blah-- has to be known. Now policy issues are obviously important. But the main issue is this one. The cost of the screening should be established.

In other words, the screening procedure, whatever it is, whether it's genotyping or tandem mass spec, has to actually be within the budget of the State of Massachusetts. And that's why, unlike 512 Genomics, they're absolutely allowed to make decisions like we're only going to look for the genotypes that account for 99% of the disease burden, because it's a public health intervention and not our doctor. And that's very, very different.

AUDIENCE: [INAUDIBLE] tests biochemical or--

ISAAC SAMUEL KOHANE: Most of those tests were biochemical. They're testing a genetic disease. But what I'm telling you is this is actually old. The cystic fibrosis test now is a DNA test. And they have it now to test that going online this is an old slide in the remaining 10 minutes. Let's roll play. Let me tell you about a disease, congenital hyperinsulinemia is the most frequent cause of severe, persistent hyperglycemia in newborn babies and children.

In most countries, it occurs in approximately one in 25,000 to one in 50,000 births. About 60% of babies with hyperinsulinemia develop hypoglycemia during that first month of life. Their blood sugar crashes. An additional 30% will be diagnosed later in the first year. And the remainder half of that. So 90% of them are diagnosed in the first year, 60% in the first month.

With early treatment and aggressive prevention of hypoglycemia, brain damage can be prevented. The brain damage is from having sustained low blood sugar, which is actually not harmful when you're adult and your brain is stable. But when you have a developing brain, and it's not getting a lot of glucose and, therefore, metabolism for periods of time, especially repeatedly, you can really have brain damage.

However, brain damage can occur in up to 50% of children with hyperinsulinism if their condition is not recognized or if treatment is ineffective in the prevention of hypoglycemia. So remember this-- one in 25,000 and one in 50,000-- on the other hand, neonatal hypoglycemia-- so hypoglycemia in these newborn kids has an incidence of two to five per 1,000. There's a zillion reasons why kids have hypoglycemia, from being slightly immature at birth to the IV was switched off too fast and so all of a sudden the pancreas didn't have enough minutes to wake up and switch off the insulin, that they're having an infection, and so on.

So the state of our knowledge in 1992, when I was finishing my residency, was there was this disease called nesidioblastosis or hyperplasia of the pancreatic islet cells, the cells that produce insulin. And for reasons that we didn't know, either the entire pancreas or spots in pancreas were hyper-producing insulin. And if we didn't treat it-- some percentage these kids would go on to be medically manageable but a whole bunch of them were not. And we'd have to take out their whole pancreas.

And sometimes we saw that we didn't need to take out the whole pancreas, that there were just focal pieces of pancreas that were involved. We could have left them more. Because by taking the whole pancreas out, we made them, A, diabetic, B, with insufficiency of those enzymes that used to dissolve your foods. It was really not a pretty sight. Plus, we kept them in the hospital for weeks to diagnose them. That was the state-of-the-art when I was a Fellow.

Here's the state of our knowledge in 2004. There is something called a potassium channel, of which there are many, which controls insulin release in the islet cells. Basically, there are these two proteins-- SUR and Kir6.2, which actually gate the flow of potassium. The channel determines the resting membrane potential, which is maintained at the necessary voltage to keep voltage dependent calcium channels closed in a cell that does not secrete.

When glucose comes into the cell and there's a resulting change in the ratio of ATP to ADP because of metabolism, K channels close the membrane that depolarizes. Subsequently, voltage-gated calcium channels are open, initiating insulin secondary cascade. Therefore, the K channel-- the potassium channel-- functions that are linked to the metabolic state of the cell and the electrical activity of the membrane, resulting in the stimulation or inhibition of insulin release.

So we have a thermostat, rheostat, which says, essentially, the more glucose I see, the more I'm going to release calcium and, therefore, insulin into the blood. So that's the basic rheostat. And we know that these two genes, which actually happen to be next to each other on chromosome 11-- if you have mutations in them, instead of coming nicely together in this octamer, they come in these various dysfunctional or poorly functional heterodimers or homooctamers. They just come together in the wrong way. And they don't work right.

And it turns out, there's a lot of mutations in SUR, one of those genes. In the Kir6.2, there's only three mutations. But looks like maybe 30, 40 mutations in SUR. Some of them are common. They're hotspots. A lot of different families have them. Some of them are one-of-a-kind. Some of them are in the coding regions. These are the 39 exons of SUR. And, by the way, that's really going to cost us in our 512 Genomics company to sequence all of those.

And some of them are in introns. They're at the splice junction. They're just right across the intron, exon boundary into the part of the intron, which is the donor or splice acceptor site, which determines when you're going to splice or not. And these all are known highly-penetrant causes of disease. They're autosomal recessive. They are autosomal recessive, but if you look at these two forms of hyperinsulinemia-- so if you look at the slide, there's two patients that have the two different presentations I told you about.

One has these focal hyperplasia, which pushes aside the normal tissue, and the other where it diffuses throughout the tissue. We now know why that is. It turns out that there is in the diffuse form, it's germline transmission of the homozygous state. And the focal state-- what's happening is there is loss of heterozygosity so that you have a parental gene-- I think actually a paternal gene. And there's a loss of heterozygosity, so it would be a loss of the maternal allele.

And so in those cells with a loss of maternal allele get focal hyperplasia due to the poor functioning of that channel. So I told you about this disease. And in the last five remaining minutes, would you order a genetic test? How about hypoglycemia in a newborn? Maybe. Jose says no. Dr. 512, are you going to order this test or not?

At least explain like a doctor because you're actually making a decision. Because we have a patient. We need a decision.

AUDIENCE: This is one in a 1,000?

ISAAC SAMUEL KOHANE: What?

AUDIENCE: You said one in 1,000?

ISAAC SAMUEL KOHANE: I think I said two to 5,000 of routine hypoglycemia. And I think I said one in 25,000 for persistent hyperinsulinemia. Yes? No? What are we going to do? Dr. Wolff?

AUDIENCE: Yes.

ISAAC SAMUEL KOHANE: Yes. So there are people like who will do it. But, thankfully, we don't give them the opportunity.

AUDIENCE: [INAUDIBLE]

ISAAC SAMUEL KOHANE: Because 512 Genomics would be incredibly happy with you because we'd be making a mint. But probably you want to look for more signs and symptoms to make you think that this is happening. So my take on it would be if there's a persistent drop in glucose, if the IV runs-- I should say less than twice the basal amines. Let me give you the full scenario.

You have a kid that you're maintaining a blood sugar by giving twice the normal amount of glucose. And every time you try to pull it down, the kid gets hypoglycemic. And it doesn't happen once. It happens many times over the first few days. And so, basically, convinced yourself that there's something wrong. So you give yourself two or three days to really convince yourself there's something wrong.

And when you do that, well-- I'm just telling you by experience-- probably you want to do that within the first week. So what would you order?

AUDIENCE: Sequences that--

ISAAC SAMUEL KOHANE: By the way, don't feel bad about it. If I asked any group of medical students or endocrine Fellows, they would not know the answer.

AUDIENCE: Sequence the genes.

ISAAC SAMUEL KOHANE: Which gene?

AUDIENCE: Both.

ISAAC SAMUEL KOHANE: Both, OK. Which part of the gene? Remember that picture of SUR. I thought we saw things everywhere, right?

AUDIENCE: Yeah. [INAUDIBLE]

ISAAC SAMUEL KOHANE: From what sample?

AUDIENCE: [INAUDIBLE]

ISAAC SAMUEL KOHANE: You could. And eventually, three months out, if we have no better story. But parents are going to come wailing at you saying--

AUDIENCE: Presumably-- [INAUDIBLE]

ISAAC SAMUEL KOHANE: No, but maybe it's not. I mean, I'm telling you that there's this-- these parents are wailing on you and saying-- and you're saying three months. I thought I came to Harvard. Don't you guys have genetic testing? So what are you going to do?

AUDIENCE: Probably just stick a needle in your baby [INAUDIBLE]

ISAAC SAMUEL KOHANE: You brute. So here's what I would do. If you ever come certainly to pancreatic removal, which we will have to, in some cases. Definitely won't want to look at the mutation tissue. But what I'll do is I'll actually look at the parents and see if a father or mother also have the mutation. So if the father, for instance, has the mutation in one gene-- if he's heterozygous for the mutation-- I have a very high index of suspicion. If the baby is homozygous, obviously-- so I look at baby blood and look at dad blood is what I would do. I'm pointing out it's not obvious. And every story is slightly different. And that's why this whole area of genomic medicine is incredibly fraught with a knowledge management problem.

AUDIENCE: This may be a really stupid and [INAUDIBLE] question.

ISAAC SAMUEL KOHANE: No, no such thing.

AUDIENCE: Since, in this particular example, you [INAUDIBLE] possibly inherited gene, why wouldn't you try to build up-- get a sufficient cell sample, extract an array, [INAUDIBLE] and just basically test to see which one those guys [INAUDIBLE]

ISAAC SAMUEL KOHANE: Are we looking expression or are we looking at DNA?

AUDIENCE: Well, we've been working on expression.

ISAAC SAMUEL KOHANE: So those were point mutations. Those were point mutations. Some of them were deletions. But most of them-- in fact, every one I showed, there was a microdeletion, three or four bases gone. Every array that I showed you would actually still say the gene was present.

AUDIENCE: But if it was in one of the [INAUDIBLE]

ISAAC SAMUEL KOHANE: No, that just changes the splicing. Or it's causing early termination of the gene product. Let's say for the sake of argument-- let's go back.

AUDIENCE: Yeah, maybe I just [INAUDIBLE]

ISAAC SAMUEL KOHANE: Let's say that it's here. And let's say that you're missing this. That you don't, in fact, have it spliced correctly. And that leads to continued translation of the intron as an exon. And, therefore, since it's mumbo jumbo, it causes premature termination. You still have 90% of the gene there that's being transcribed. And that RNA, therefore, is going to be registered by the expression array.

Now you could say maybe I want to have a resequencing array. We still have to resequence the gene, no way around that. OK, where would you send it? We know the answer. We'd look up gene clinics, gene tests. What would you tell the parents when you're obtaining of the consent? Well, I'm running out of time.

But I think in addition to all the things that we said before, we actually have to tell them that we might not find the cause of it. But that if the father's shows up one thing, that might be one result. And therefore, we may only have to hack out a bit of the pancreas. If the kid's homozygous for it, we may have to take out the whole pancreas and so on. So that's it for today. That's really the state-of-the-art of genomic medicine, 2004.