SPEAKER: So I think well go ahead and get started. Welcome. Thank you for coming to attend the CCaTS Grand Rounds. In my role as director of drug discovery for the CTSA it gives me great pleasure to introduce today's speaker, who is working on the application of pharmacogenomics to novel drug therapies. So Dr. Pereira is a good friend and colleague. And he did his medical school at the University of Bombay. Thereafter he went for internship and residency at Connecticut, fellowship at Brown, and then second fellowship in heart failure and cardiac transplantation at Harvard. After he graduated from Harvard, he went into a private practice, where he had a very successful private practice at USC. But to his great credit, he decided he wanted more of an academic challenge. So he left USC in 2006 and came to Mayo.

When he came to Mayo, he was a regular clinician, had no dedicated research time. But in his own time, he carved out a research niche and has been very productive in that regard. Since being here, he's published 62 peer reviewed papers in high impact journals, such as *Fassive* and *Circulation,* in addition to which, he was the recipient of one of our KL2 scholar awards. And since then, he's successfully applied for and received a co-investigator award and a UO1. And he was just telling me, he has two other applications that are going to be reviewed next week. So together our fingers are crossed for him to Naveen.

So, Naveen, thank you for coming to present. We look forward to your presentation. And congratulations on your stunning success so far.

[APPLAUSE]

NAVEEN PEREIRA: Thanks. Thanks, Andy. You've kind of been along with me this entire journey. And I appreciate the support, appreciate the support of the CTSA and the entire committee, and, of course, my mentors for the KL2 award. So today I'm going to discuss, really, the concepts of cardiovascular pharmacogenomics and how I've come to train myself in this field. And these are the disclosures. I personally have no disclosures. And we are going to be discussing the use of Ticagrelor as an off-label use in a clinical trial that I'll talk about towards the end of the talk.

So the objectives of this talk is to really recognize the concept of how genetic variation can affect drug response. We want to identify the strategies that we've used in the laboratory and also the population level, as to how genetic variation should be investigated in terms of looking at phenotype or drug response. And then we'll review the implementation of pharmacogenetics in clinical practice and then discuss the future direction that our cardiovascular pharmacogenomics program is going to be heading towards.

So here is the sequence explosion, as you may term it. So since the human genome was unraveled in 2001, the cost of sequencing has dramatically decreased. It used to be about $10,000 for a million base pairs. And now the third-generation sequencing, we are about $1 for a million base pairs. So you can get your whole genome sequence for about $2,000 to $3,000.

And with that, the amount of information that's available in publicly available archives has exploded. There are terabytes and terabytes of information that are available. So with this information, people have used techniques like genome-wide association analyzes to try and correlate disease with genetic variation. And in 2005, one of the first genome-wide association studies showed the correlation of Crohn's disease with a genetic variant on chromosome 1.

And I took this from the NHGRI catalog. And can see there's been a plethora of genome-wide association studies just over seven years, with the decreased costs and more educated and interested investigators trying to figure out the association of genetic variation with different disease and other drug treatment response phenotypes. So with this knowledge, have we become like the blind seer of thieves Tiresias.

Do we have this knowledge? Can we do something about it? Or are cursed where we just have this knowledge and do nothing? And I think that's why pharmacogenomics is so interesting as a field, that if we are aware of what role inheritance plays in individual variation in drug response phenotypes, we can at least attenuate the way we treat our patients.

And the clinical goal is really to avoid adverse drug reactions, to maximize drug efficacy, to really select the right patient. But there are also some really important research goals that I've learned over the past several years. We can discover new pathways, understand new mechanisms using the wonderful techniques in the laboratory.

So the heart of the research strategy is what is the relationship of the drug on the clinical phenotype? And at the center of this premise is genetic variation of single nucleotide polymorphisms. And to validate that indeed this relationship exists, you really need to replicate your findings. And you have to functionally validate them in the lab.

So therefore, my training under Dr. Weinshilboum's mentorship has been kind of three-tiered. So what we did was we first examined candidate genes in known pathways, looked at genetic variation, and tried to figure out what the impact of that genetic variation is in the laboratory or functional genomics as a facet or field. And then, adopting genome-wide association, take an agnostic approach, looking at genetic variation across all the chromosomes, and trying to associate it with clinical phenotype in a population, of population genetics or discovery genomics.

And then finally, when you have information of a certain genotype and you know it affects drug treatment, can you base your drug treatment based on that genotype and therefore affect outcomes? So we've gone through all three processes. And the first one I'm going to discuss is the functional genomics.

So I've been really interested in the natriuretic peptide system. We have a strong cardiorenal laboratory. And these are three natriuretic peptide receptors. And I want you to pay attention to NPR-C, NPR-3. It's a clearance receptor for three important natriuretic peptides. And what are natriuretic peptides? They're responsible for natriuresis. They control volume, blood pressure. So it's very important in hypertension, heart failure, and stroke.

So these natriuretic peptides are cleared by natriuretic peptide receptor C or 3. They are also degraded by MME, or it's also known as neutral endopeptidase or NEP, which is present in the kidneys. So as I mentioned, the natriuretic peptides and the whole natriuretic peptide system and pathway plays an important role in hypertension, stroke, and heart failure.

So my first project was to take MME, which is responsible for degradation of natriuretic peptides and resequence the MME in using DNA samples from a multi-ethnic population. So there were 96 Han Chinese, 96 caucasians, and 96 African-Americans. And the idea was to take these normal, so-called normal people, and identify polymorphisms. And then see what the functional implications of those polymorphisms is. Because identifying normal genetic variation in MME could alter natriuretic peptide metabolism, and therefore, have important cardiovascular effects.

And so here's a schematic description, depiction, of MME, the gene. You can see that 23 exons, as depicted by these boxes. And these are the three populations, the Caucasian-Americans, African-Americans, Han Chinese. The arrows really depict the polymorphisms. And the color of the arrows that depict the allele frequency. And so essentially, by using Sanger sequencing, we identified 105 polymorphisms in this gene, of which 80 were completely novel and were not described. And more importantly, there were eight nonsynonymous polymorphisms depicted by these boxes, nonsynonymous, amino acid-changing genetic variants.

And of these eight nonsynonymous polymorphisms, one was previously described. Seven were completely novel. So here is a diagrammatic description of these nonsynonymous polymorphisms in transmembraic structure of the protein. This is the extracellular domain, transmembrane domain, and the intracellular internal cytoplasmic domain. And so you can see how these nonsynonymous polymorphisms are located in this protein structure.

So what does this mean? Does amino acid change that occurs, does this variation, does it impact on protein structure and function? And so it comes to bear that you really, if you have genetic variation, great. But what are the functional implications of it? And to test the function of these genetic variants in the laboratory, we took MME expression construct and did site-directed mutagenesis and were able to recreate all these nonsynonymous polymorphisms in different varying constructs and then transfected COS-1 cells and looked at protein expression and also examined enzymatic activity.

So protein expression was done by Western blot analysis. And we did quantitative protein expression. And MME enzyme assay was done by a fluorometric assay. And these were the results. And what's interesting is here is wild-type. And wild-type is 100%. And everything is compared to wild-type. In the yellow, the protein levels, and orange is the enzymatic activity level. And you can see that all nonsynonymous polymorphisms don't necessarily affect protein expression.

And in fact, the only nonsynonymous polymorphism that affected protein expression was valine-73, a variant allozyme. And both protein expression and enzymatic activity was markedly reduced as compared to Wild Type. So with that collaborated case Western, we basically looked to see and predict is this really true? Does it contribute to protein instability?

So if you substitute methoionine-73, which is wild-type allozyme with valine-73, it generates multiple short contacts that contribute to protein instability. So by using computational structural modeling, we were able to predict that this variant indeed, as compared to the other variants, would result in decreased protein expression.

And in fact, using immunofluorescence, you can see in green here is MME. And it forms microaggregates in the variant allozyme cells. And these microaggregates indicate that there is protein misfolding that occurs. And in fact, the variants, as depicted in the orange, have higher expression of heat shock proteins, like binding amino globulin and GRP94. So here you have a structural modeling based on X-ray crystallography structure, demonstrating that amino acid substitution can lead to protein instability. That protein instability was reflected by us seeing microaggregates in the variant and then seeing higher expression of heat shock proteins.

And indeed, if you take the variant allozyme and treat it with MG132, which inhibits proteasome degradation, you will see that the variant allozyme valine-73 that is markedly reduced then increases in protein expression, therefore, reflecting that proteasomes play a really important role in this protein degradation. Similarly, treating the cells with 3-methyladenine and inhibiting autophagy increases protein expression, also reflecting the fact that autophagy plays an important role in this variant protein degradation.

So essentially, using a very similar approach, we took the other gene, natriuretic peptide receptor 3 gene, and did Sanger resequencing in this multi-ethnic population and identified about 95 polymorphisms, of which 50 were novel, and again, identified eight nonsynonymous SNPs, depicted in the boxes here, of which seven were novel. And again, we did protein expression by quantitative Western blots. And you can see, there were actually five variants here that were significantly reduced in expression as compared to wild-type. And we decided to focus on arginine-146, because it was the most markedly reduced in terms of protein quantity.

Again, doing computational structural modeling based on the X-ray crystallography structure, we see that a spare gene, when it is substituted by aspartic acid at position 146, it generates multiple short contacts and again, promotes protein instability. And when treated with 3-methyladenine, you restore protein expression, indicating the important role of autophagy. So this was all very nice and interesting in the laboratory. And then how do we translate these findings and the importance of these findings in populations?

And essentially, here's a diagrammatic depiction of a natriuretic peptide receptor 3. Here are the nonsynonymous polymorphisms. Here is the ligand AMP. And of all the eight nonsynonymous SNPs that we had sequenced in the multi-ethnic population, there was one SNP that had significantly high minor allele frequency of about 30% or so. And this particular nonsynonymous SNP was also interesting, not only because it's common, but it also interacts with a G protein that's coupled to adenylyl cyclase. And this is important because NPR3 is now considered to be an important receptor in downstream cellular proliferation signaling.

So would a nonsynonymous polymorphism affect the interaction with G protein and affect downstream signaling? And can we see that at an individual level in terms of population analysis? So here is the background. So natriuretic peptide clearance receptor NPR3 plays an important role in clearing natriuretic peptides. It can therefore affect hypertension risk. And it also can modulate smooth muscle cell and fibroblasts proliferation, and therefore, could affect cardiac structure and function.

So would a nonsynonymous SNP in that particular receptor affect natriuretic peptide levels? And can it affect cardiac structure and function? So we took a cohort of about 2,000 participants from Olmsted County and used the MetaboChip. So it's really not a genome-wide association study, but it does examine genetic variation across 16,000 genes, 200,000 SNPs. And what we found was that there were approximately 778 subjects who were either heterozygotes or homozygotes for this particular genetic variant.

And so what we did was perform an analysis comparing the homozygotes with the wild-type and the heterozygotes, so using a recessive model. And in a sense, the most significant difference between the carriers, or the homozygotes versus is the heterozygotes and wild-type, was that diastolic dysfunction was significantly different. So homozygotes had significantly increased prevalence of diastolic dysfunction and a prolonged deceleration time on echocardiography, reflecting there were relaxation abnormalities within the myocardium. And this is just a depiction of the increased prevalence.

But what was interesting is when you fit the genotype in the multi-variable model, and you take important variables that affect diastolic dysfunction, like age, female sex, hypertension, BMI, the genotype remains significant. So this was actually the first paper to indicate, in the diastolic heart failure community, that NPR3 can play an important role in diastolic function of the heart. No one else had pointed this out before. So here is using genetic techniques, looking at the phenotype and the significance of the phenotype, initially starting from our resequencing experiments and then seeing what role it plays in the population.

And interestingly enough, the homozygotes, despite there being a nonsynonymous SNP in the receptor, did not have any different natriuretic peptide levels as compared to the wild-type or heterozygotes. And this indicating that this is nonsynonymous polymorphism did not affect the clearance function of the receptor. And it most likely indicates that it affects the downstream signaling pathways of the receptors.

So what I showed you was examples of taking specific genes, looking, examining the impact of the variance in the laboratory, and then looking at its impact in the population, and seeing what the phenotype is. And so the second approach is taking agnostic perspective and looking cross the genome, using genome-wide association techniques, but in this case using the MetaboChip, and trying to discover new variants associated with a phenotype of interest. So one phenotype that I was very interested in is circulating atrial natriuretic peptide levels.

We know-- and there was a nice nutrigenetics paper published, which shows that reduction in ANP levels predisposes people to hypertension. And as we know, hypertension is very common. It's also predisposes to development of heart failure. So we also know that the variability in natriuretic peptides is, in part, accounted for by genetic factors. So no one before had really tried to identify what are the genomic variants that contribute to the variability in atrial natriuretic peptides? We're asking a very important question. What are the genetic determinants of variability in ANP? ANP plays an important role in hypertension and in heart failure.

So again, it's the same population, parrot population, which is part of the REP, used the MetaboChip. And this time what we did was we-- it's very important, as I described before, that to functionally validate, to validate your SNPs, you have to have a replication cohort. So we randomly assigned 891 subjects in a replication. We use 893 for discovery. The ANP levels were measured using a radioimmunoassay.

And this is a Manhattan plot. And the chromosomes depicted on the x-axis here-- each skyscraper is a chromosome. All of these dots represent genetic variants. And the y-axis is the logarithmic p-value. And any p-value greater than 10 to the power minus 7 was significant. And you can see, depicted in red here, were all of the significant genetic variants that were associated with ANP levels.

And so it goes back to replicating. So you find that in the discovery cohort. And I don't expect you to pay attention to all these numbers. But I do want you to see that there is remarkable consistency.

So here are the variants. Here are the genes those variants are in. Here is the effect on ANP levels in terms of fold change. And here are the p-values. And there was remarkable consistency for all these variants in terms of replication. And the direction of fold change was similar, really lending validity to these SNPs playing an important role in determining variability associated with ANP.

This is a LocusZoom plot. And you can see most of these significant SNPs, as depicted in red and green, are located in this gene cluster, MTHFR, CLCN6, Nppa and Nppb. So they are all in tight linkage to cyc-librium. So they kind of go hand-in-hand with each other. So it's hard to sort out any one SNP that is implicated with a phenotype.

And so this is a LD plot. And You can see all of those SNPs are in tight linkage to cyc-librium. And so what we did was take the SNP RS5063 because it was the biologically the most plausible SNP. It's a nonsynonymous SNP in Nppa, the gene that encodes ANP. So we said, why not take that that? It represents most of the genetic variation within the region. And then we decided to look at the functional effect of that variant in the population by looking to see what the phenotype was of the carriers of that particular gene variant.

And you can see right off the bat, indeed, the carriers of the minor allele, both heterozygotes and homozygotes, had a significantly higher diastolic pressure, blood pressure. They had a trend towards increased coronary artery disease, and decreased HDL levels. So there was metabolic syndrome type of picture. And this made perfect sense because the ANP levels was markedly reduced and the carriers of the minor allele as compared to the wild-type. And there was a high prevalence-- trend towards a high prevalence of lefant ventricle hypertrophy. And there was some cardiac structural modeling effects.

And so here are the carriers of this genotype, which was significantly accounted for the variability in ANP during this genotype-phenotype association study. And those carriers had a markedly reduced ANP levels. And remember, ANP controls your blood pressure. So those carriers had a higher blood pressure, more left ventricle hypertrophy, abnormal hearts.

And so what does that translate? And the nice thing about this cohort is we have about 10 to 12 years of followup being part of the REP. So do these carriers of this genotype, with having low ANP levels, some metabolic abnormalities, and higher blood pressures, does that translate to something? And what we found was that the carriers of this minor allele have a higher risk for stroke.

But you may say, well, it's all the blood pressure and the metabolic abnormalities that may have contributed to the higher risk of stroke. And you can see in a multi-variable regression model, you take important variables like age, female sex, diabetes, hypertension, atrial arrhythmias, cholesterol level, the genotype still remains significant. And the hazard ratio was approximately 1.6 in determining the risk for stroke.

So this is, again, for the first time showing a nice link between what genetic determinants are responsible for variability in ANP level, taking the minor allele, the carriers of minor allele, seeing what their phenotype is, and seeing what the outcome is, and getting all those things together. So finally, besides exploring genetic variation in the laboratory and the population level, this is what we do. We are physicians. So how can we apply from the bench to the bedside and translational pharmacogenetics?

So there are significant challenges. So when you have a pharmacogenetic or genomic discovery, to get into practice there are challenges. And the challenges come from what is the evidence of clinical utility? Is the incremental value beyond the current testing? Like for example, warfarin, does measuring, assaying the genotype, is that more important than measuring INR?

What do the practice guidelines say? Is this genotyping available in CLIA-approved environment to be used in the medical record? How do we incorporate it in medical records? How do we create alerts for physicians? Is there insurance coverage? And finally, what do patients and physicians feel about genetic testing?

A good example is-- and my mentor discovered this particular variance in TPMT and how it can impact on metabolism of azathioprine. So you get a loss of function alleles in TPMT. And if you are a carrier for a loss of function allele in TPMT, you are not able to metabolize 6-mercaptopurine. And so you are a higher risk for toxicity with azathioprine. So azathioprine decreases your white blood cell count. So if you carry this variant, and you're homozygote for it, you can get life-threatening myelosuppression because you cannot degrade mercaptopurine.

So we decided to look in a heart transplant population. What does this really translate when we do it clinically? And so we didn't have any homozygotes, because it's well established in homozygotes that this is a problem in terms of toxicity. But we had heterozygotes, about 20 plus heterozygotes versus 70 wild-types. And we saw no difference in leukopenia for the heterozygotes. But paradoxically-- now since we have more azathioprine, you would think that the heart transplant patients would have less rejection. And paradoxically, we found increased rejection and increased risk for discontinuation of azathioprine in the heterozygotes as compared to the wild-type carriers.

So sometimes what we think in the laboratory or logically makes sense, when you actually translate into clinical practice may not pan out. And so therefore, there is some element of skepticism. And a good example of this is clopidogrel pharmacogenetics. And clopidogrel is a very important cardiovascular drug. There are about three million prescriptions written every month. The 2010 sales were in billions of dollars. And it's indicated as an antiplatelet agent for myocardial infarction, CVA, and after PCI.

So clopidogrel is a pro-drug. And it gets converted to an active metabolite by all these cytochrome P450 enzymes. But the enzyme that's most important for this biotransformation is CYP2C19. So you need a CYP2C19 along with the other cytochrome P450 enzymes to have the active metabolite of clopidogrel, which can then irreversibly bind to platelets and inhibit platelet aggregation. And so the most important variants, and the most common variant, is *2. And carriers of *2, as you can see the minor allele frequency in Europeans is 15%, but very common in Asians.

So these are the loss of function alleles. So the carriers of *2 and *3 are unable to convert the inactive drug to an active drug. So clopidogrel should "not work," quote-unquote, in carriers of *2, *3. Three *17 is felt to be a variant that results in increased conversion of the active metabolite. And there's some controversy about it. So we are not going to discuss that today.

So you can see there have been multiple retrospective studies done. So these are multiple clinical trials with acute coronary syndrome and after PCI. And the carriers of *2, *3, reduced function alleles for CYP2C19 have a higher risk for stroke, myocardial infarction, and death. And based on these multiple studies, the FDA placed a warning in the clopidogrel or Plavix drug prescribing information, saying that effectiveness of Plavix depends on conversion of the cytochrome P450 enzyme. Poor metabolizers have a higher cardiovascular event rate. There are tests available to find out the genotype of those poor metabolizers. And consider alternative treatment.

So you would think all us cardiologists would be rushing to genotype our patients prior to prescribing clopidogrel. And so as soon as that FDA warning came out, there was an ACCHA panel that was put together to respond to it in a formal way and to guide the cardiovascular community. The first author of which was David Holmes, who is the president of the ACC at that time and is a faculty member of cardiology. And the bottom line was, they said the evidence base was insufficient to recommend either routine genetic testing. And there's no information that such testing would improve outcomes, OK, which is true. Because most of these studies were retrospective and observational.

The press was following the story very closely. They said, well, Plavix may not work if you have a certain genotype. And then there was a study, metanalysis published, that says, actually, these patients may not be at an increased risk. So the study challenge the FDA and Plavix. And now Plavix is generic. So it's about one sixth or one eighth the cost of the patented drugs that are available, antiplatelet drugs. So because one solution would be give everybody the newer antiplatelet drugs that don't depend, don't get metabolized with the CYP2C19 19 pathway. That's all well and good. But the problem is there's increased bleeding with those medications. And they are six to eight times more expensive now with it going generic.

So we are still confronted with this black box warning. And we don't know whether altering therapy based on this genotype can affect clinical outcomes. And so we put together a TAILOR-PCI trial. It's an innovative trial to address the implementation of genotyping and, based on the genotype, tailor the antiplatelet therapy after coronary stenting of PCI in the catheterization laboratory.

And so here is the study design. So the patients who undergo PCI-- and you know coronary stenting. There are about a million procedures performed every year in the United States. So here are patients who undergo PCI. And they get randomized to what we, 99% of cath labs do around the world. They just get clopidogrel 75 milligrams a day. They get no genotyping. And the other arm, we decided to test prospective genotyping.

So we partnered with Spartan Biosciences, who are able to provide us with the results within 45 minutes. So within 45 minutes of the patient getting randomized and consented, we can tell if the patient is a carrier of *2, *3, the reduced function allele. So then we follow FDA guidelines. So if you're a *2, *3 allele person, you get ticagrelor, the alternative drug. If your wild-type, you get clopidogrel.

Now this other arm, we don't know really what genotype they are. And we had a discussion with the FDA that why can't we, up front here, genotype everybody, take the *2, *3 allele carriers and randomized them to ticagrelor or clopidogrel? And they said, if you know the genotype, you will treat them with alternative antiplatelet drugs. So we decided to be ostriches and bury our head in the sand and not know the genotype until one year, when we'll do retrospective genotyping, using TaqMan assay. And then we'll know who are the carriers of the reduced function CYP2C19 allele. And then we'll compare the outcomes of *2, *3 CYP2C19 allele carriers who are treated the clopidogrel versus those treated with Ticagrelor and look at hard end points of myocardial infarction, stroke, and that and see if there is any difference.

5,300 patients, we've got 16 participating centers around the world. There are three in Korea. There are four in Canada. And the rest, nine centers are in the United States. All the Mayo centers, Mayo Clinic, Rochester, Arizona, Florida, and the two health systems centers in La Crosse and Eau Claire are participating. It's really amazing, the partnership, because this is a Mayo-funded study finale. And the reimbursement per patients is very low.

This was possible, really, in an altruistic sense. The PIs have been participating. It's an innovative trial design to address FDA regulatory issues. We're using really state-of-the-art, point-of-care almost genotyping platform. And we've randomized close to 700 patients so far. So the trial is up and running and doing well.

So what are future directions? How do I integrate all of this knowledge that I've acquired over the past several years and use it as we move along? So I've put together an interesting population of patients with recent onset dilated cardiomyopathy. And these are patients from an NHLBI-sponsored trial called IMAC-2.

There is a German cohort. There is a Czech cohort. There is an Italian cohort. There are about 1,000 patients. And we're going to do genome by association analysis, a pathway-based gene analysis, and hopefully identify candidate genes, candidate SNPs.

And really, what I want to point out here is to functionally validate those genes, the SNPs, we are going to use iPS cardiomyocytes derived from iPS cells. This is a great resource. It's an example of using SIM and regenerative medicine and combining resources to really answer an important question, what accounts for variability in treatment response for heart failure? And that's my clinical expertise. And so it's very dear to me.

And at a acellular level, we can look at cardiac myocytes derived from iPS cells and look at hypertrophy. So these iPS-derived cardiac myoctyes, you can see that the sarcomeres are very linear. But you treat them with norepinephrine, which is an accepted model, to create hypertrophy. You can see punctate densities here in the cardiac myocyte. And then you can compare.

So usually cells that are hypertrophied have these disarray of the linear architecture of the sarcomeres. And interestingly enough, if you treat with metoprolol, you can see that the percentage of disorganized cells decreases. So here is a great phenotype. You can use cardiac myocytes derived from iPS cells. The advantage is we'll know the genotype. And therefore, we can take the genetic signal from GWAS, take that specific genotype in iPS cells, and test it using accepted methods of hypertrophy and treatment, and validate our observations in the laboratory.

So to summarize, functional genomics, which I learned in the laboratory of Dr. Weinshilboum you can elucidate the impact of genetic variation by using in-vitro mammalian cell systems. Candidate gene or GWAS findings can help us define pathophysiology of disease. The translation of pharmacogenetic discoveries to clinical practice remains challenging. We have to engage in large clinical trials, like we are doing, to really impact clinical practice. And the future lies in using innovative models like iPS-based models to explore new pathways.

So this is the guy, who I think-- and this is a saying by William Ward, where he says "mediocre teacher tells. Good teacher explains. Superior teacher demonstrates. But the great teacher inspires." So this is the man who has inspired me, taken a clinician and tried hard-- it's been an uphill battle-- but tried hard to make into a scientist. And I really owe everything to Dr. Weinshilboum. He's been a fantastic mentor.

And obviously, this would not have been possible without the support of my chair, who has protected my time, and by other KL2 mentors, Dr. Redfield, Sue Bielinski. John Burnett's been a great collaborator. Vivian Yee did all the computational structural modeling. A huge TAILOR PCI team, literally hundreds of people involved in the trial-- in the functional genomics laboratory, Dr. Wang, who always puts me straight and gets me focused on doing the right thing; Pinar and Dong Lin, who helped me with the functional genomics work that I showed you; and Linda Pellymounter, Irene Moon, who helped us with resequencing; biostatistics support, and my new collaborators and mentors, Tim Nelson, Tim Olson from regenerative medicine. We're going to use medical informatics for replication of the GWAS in this last study I showed you, so Chris Chute, Jyoti Pathak, and Les Cooper.

And with that, I want to say thank you for your attention. I really appreciate you being here today.

[APPLAUSE]