I have to say, while we're starting, I mean I'm reading some of the chats going on back and forth about some of the weather, and I think me and my colleague in San Diego are both in the same spot, we're not sunny and 80, but we're somewhat partly cloudy and in the 70s. So I will say to you, this is really -- this part of the presentation is, you know, supported by my governor of Florida for our tourism, so it's a great place to be right now. But even what makes it better is that we have Chuck Peloquin here in Florida.

And I don't know about you guys, but, you know, for the longest period of time, you know, this whole concept of doing drug levels had been somewhat confusing, and it's kind of confusing in medicine, you know, when you look at other diseases, doing drug levels for antibiotics or other therapeutic drugs, it's a natural. I mean you do it all the time. And what always kind of interested me is that for tuberculosis, no matter how long we've had these antibiotics, and as you know, we've had TB antibiotics almost longer than almost any other antibiotic, and despite that, we've always used the approach that one-size-fits all, you know, everybody 300 milligrams of INH, everybody gets Rifampin 600 milligrams, and in some miraculous way, right, it seems to work. And that's where we're really, really lucky.

What the bottom line comes down to is I think all of have been in a position where we know that one size-doesn't always fit all, you know. And this has, I think, become even more of an issue recently. I think we would agree that as we're seeing TB spread into other populations that have other conditions, such as immunosuppressive diseases or they have organ failures or they have kidney failure or liver failure, or if these patients are on public drugs that interfere with the absorption, or even worse now is that when we're looking at cases, like, multi-drug resistance or XDR cases, where we're using drugs where the absorption is not quite the same, we're definitely in a situation now where we kind of know that we kind of need drug levels, you know. But here's the part where, while we kind of feel we need drug levels, it's a visceral feeling, most of us don't know quite know exactly when we should get it or how we should get it, and then worse yet, we don't really know what it means when we get them back. And that's where Chuck comes in.

You know, I don't know about you guys, but, you know, in our world, you know, in our life, there are some people that are just known by one name. Everybody knows -- if you say Madonna, everybody knows Madonna, you know. Or if somebody turns around and says Beiber, you know who Beiber is; right? But you know, in TB we have that person with one name, and his name is Chuck. And I met Chuck Peloquin in 1993. I went to National Jewish to do my training, and I was starting out, and I remember being struck by Chuck's lectures on therapeutic drug monitoring. And at the time, I was take over A. G. Holley and I had a hospital filled with patients who were just failing, and they weren't getting any better. And even despite using the one-size-fits-all approach, these patients failed. And the bottom line was, it was up to me to try to figure out how I was going to cure them.

And you know what, I got to say I really do mean this, Chuck turned out to be one of those people who really influenced me, and he was the one who really said that, you know, many of these patients may be having problems with absorbing their meds or interactions with their meds, and Chuck was a hundred percent right, you know. And since that meeting in 1993, I'm really proud to say that Chuck and I have, you know, been friends. I'm really proud to call Chuck a friend, though I would never admit it in front of him. You know, as you guys know, friends have people's backs, and Chuck definitely has had my back. And over the years, I can't even begin to tell you how many times Chuck has saved my back and has helped us cure these patients. Of course, I take all the credit for the cure. Chuck gets none of it.

But I really think that there are certain cases and situations where drug levels become so important, and I think what is interesting to me is that lecture, believe me I would never ask or request Chuck to do a lecture, because, look, you know, I'm tired of him. I just don't want to hear him anymore. But, you know, you guys actually were the ones who asked for the lecture, so anything that happens it's you. But the reason you're asking for it is because you all are being confronted with more and more difficult cases. Drug levels are more and more becoming the answer, and that's why I'm really proud to say that now, three years ago Chuck came down to the University of Florida. I'm proud to call Chuck not only a friend but a colleague.

And as you know, Chuck is Professor of Pharmacology at The University of Florida. He's the director of the Infectious Disease Pharmacokinetics Lab, and most importantly, Chuck is Chuck. So without further ado, I'm really proud to introduce the Chuck, Chuck Peloquin. Chuck, it's all yours.

Well thank you, David, but in all fairness, I also have to give a shout out to Chuck Daley In Denver. He also works in TB a lot.

Chuck, he doesn't compare to you. You take the Chuck. He's Chuck two.

Well we had the thing while I was working in Denver that I was original Chuck and he was new Chuck. So with that introduction, what we'll do today is what I'd like to do is take a little bit of time with each of the slides and spend some time discussing the concepts on them, and while some of the slides might have been presented in other settings, I'm putting them together here so that, from start to finish, you should have a reasonably good idea about what we're doing and why we're doing it. So let's see, yes, the control buttons are working on the screen, so that's always a good thing.

So what's the big idea anyways? Why are we even bothering with this? As David mentioned in his introductory comments, there are basically two ways to dose a drug. You can guess or, in other words, just give everybody the same dose, and we'll make the assumption that that's the right dose for everybody. Alternatively we can use what's now being called "personalized medicine." It's had different names over the years, and the meaning has expanded to include new tools, but nevertheless, instead of guessing at the right dose, we can find the right dose for each individual.

Now in the modern sense personalized medicine is generally referring to individualized drug therapy that includes genetic information. And you use that information to see if a patient is able to metabolize a drug or not able to metabolize a drug. If you're giving a really, really toxic drug such as a cancer chemotherapeutic agent, you might want to know that information up front. I'm just going to pause here for a moment. I am hearing some background noises, so if everybody could just double check that their phone is muted, that would be great.

So back to the presentation. At the bottom of the slide we see a little diagram of DNA. And if you have enough DNA, you can have genes after all. Now in the old-school sense, personalized medicine refers to what we're going to talk about today, therapeutic drug monitoring, and that, in other words, is seeing how much drug actually made it into the patient's blood and seeing how long it hangs out there. So what we want to do is take a look at each individual patient and see what they do with the drug.

So, given my age, I'm an old-school kind of guy. I get my weather report by standing outside. I guess we just fast forwarded on the slides, so let's see if we can go back. And apparently it's not letting me go back. Anyway, there was a Yogi Berra quote there that said, "You can observe a lot just by watching." So it looks like the slides are auto-forwarding on me. That's pretty interesting. So if Lolia can jump in and halt the advance of the slides, that would be great. Yeah, I think that's where we want to be. There, right there, beautiful. So we're having an interesting little event. Maybe it's the cold weather affecting the transmission, but the slides seem to be auto-forwarding.

Chuck, I think I might have fixed that. Is this the right screen now?

Okay, here we go. So where we left off was the famous quote from Yogi Berra, "You can observe a lot just by watching," so that's what we do. We take a look at what's going on inside the patient's circulatory system. Now TB treatment is guideline driven, and that, for the most part, is a very good thing. Now this is an excerpt from the 2003 guidelines. Just as an aside, David and I are working with a group of people that are formulating the new guidelines, and that's something that's happening right now. But if we look at this guideline, I'll just point up a couple of things.

If you look at the third column under "Adults and children," it shows, say for Isoniazid, there's adults and then it says "maximum," afterwards, and in children “maximum.” And, hopefully, in the new guidelines that's going to be removed, because most of the studies that were done with these drugs, everybody got

the same dose. So you could say just as easily that it's the minimum dose or the usual dose or, if you look at the study, it was the only dose. The problem I have with “maximum dose” is it implies that if you go above this, something bad might happen, and that is often not the case, and if you prove that a patient has low concentrations, despite a standard dose, then they may need a higher than usual dose, and that's why that term at "maximum" is a little bit confusing.

Now another thing that we hear from time to time is, well, Chuck, there's no prospective randomized trial specifically of therapeutic drug monitoring as an intervention in tuberculosis, and that's true. There's a lot of circumstantial evidence, but there's not that kind of prospective trial. But that's also true for a large number of items inside the guidelines. So if we just look further over under dosage you see a 2X-times weekly regimen, and that's the so-called "Denver regimen," and it's been shown to work. But if you go back to the original study that was published by Dave Cowen and Company, back in 1990, there were 125 valuable patients. So while it was a nice study, it's not the biggest study that was ever done, and nevertheless, that's one of the choices that's listed in the guidelines.

So I just point up here that even though we may not have the definitive data on prospective randomized trials for this technique, I think as David pointed out for a lot of other drugs the data are also are not fantastic, whether they're seizure drugs or anti-rejection drug or other antibiotics, nevertheless, it's a useful tool.

So guidelines are very, very important. I want to stress that. And they're very useful and they're the right place to start. Once you start treating a patient, things don't always go as planned and so what we're going to talk about is "and then." The picture there is from the movie "Dude, Where's My Car," and there's a scene in there about "and then." I've not seen the whole movie, but I've seen the clip on YouTube, and it's pretty funny.

So when we look add the guidelines, there's an implicit assumption within those guidelines that if you get the patient to take the drugs they will be cured. And this is based on a number of studies, largely done, but not exclusively done by the British Medical Research Council. But if you look at those trials, those were all per protocol statistical analyses. So in other words, when they published the paper, they only published the patients who actually were able to stay on the regimen as originally designed. And the dropout rate from those studies might have been 5%, might have been 20%, and some of the other studies, not necessarily the BMRC study, the dropout rate is over 40%. And, unfortunately, in your clinic, you still have to take care of those patients, so that's the difference between a per-protocol data analysis, which typically was done back when those studies were conducted in the 1960s, '70, and '80s.

Now a lot of studies are published with an intent to treat or a modified intent to treat analysis. The difference, of course, being that you have to account for all the dropouts, and they may be considered as failures for any number of reasons. So while we often say, well TB regimens are more than 95% effective, it's important to keep in mind that that really was a per-protocol analysis and didn't include all the people who dropped out.

So, given that, and given the new challenge that is David alluded to -- people who are on immuno suppressants, people who have HIV, people who have diabetes or renal failure -- that we're treating with tuberculosis, these assumptions are being challenged. There was a recent paper by Tawanda Gumbo and his group out of Texas, and this is a study that was entitled "Multidrug Resistant Tuberculosis Not Due to Compliance but to Between Patient Pharmacokinetic Variability," and I'm referring to that paper to see exactly what they did.

Now this is a hollow fiber model experiment, so it's an in vitro experiment. And you might say, "Well, Chuck, I don't treat hollow fiber chambers in my clinic, I treat the patient," and that's true. The nice thing about these kinds of models, though, is that you can isolate the activity of the drug from all the other activity in the system. So the interaction between the host and the organism, between the organism and anything else in the body, as far as the cavitary lesions, how big the lesions are, how long they have had the disease, all of that can be separated out. Here it's just sort of a one-on-one drug versus bug. And

that's the beauty of these kinds of models. They can allow you to look at the actual pharmacodynamics of the drug just against the bug.

Another paper by Tawanda is a recent paper, and it's entitled, "integrating Drug Concentrations in Minimum Inhibitory Concentrations" or MICs, "with Bayesian Dose Optimization" and that's a computer program approach to using serum concentrations and using that for multidrug resistant tuberculosis. And, again, I'd refer you to that paper in the " European Respiratory Journal" to see what the authors are talking about.

You will see a paper further on in the program today by Roger Jelliffe, who talks about the exact same things, but in a more general sense, and that paper is from 2000. And then finally, a paper that I published back in 2002, "Therapeutic Drug Monitoring in the Treatment of Tuberculosis" There's a new version of this it. It's being reviewed at the same journal of drugs, and we'll see when that will be forthcoming. But nevertheless, this older paper still contains a lot of the information that we utilize currently, because the drugs really haven't changed. And it explains and approach to handling the data that you might have if you get serum concentrations for your patients.

So standardized doses would be like getting a ticket to a Gator football game that didn't tell you the section, the row, or the seat, and you show up at the game and the usher says, "Okay, there's one empty seat left and it's your seat, and it's somewhere in there, good luck." And it wouldn't be very specific to what you wanted, and that is to get in your seat as soon as you can to watch the game. And, hopefully, the Gators would win, but that's another story; right.

What pharmacokinetic monitoring allows you to do is know exactly where you seat is. So it might be under the "G" for Gators. So this might be your seat right here. Let's see if I can get the tool to work. There we go. So this might be your seat right here under the "G" for Gators, and essentially this is an individualized seat, and what we're doing with therapeutic drug monitoring is getting individualized concentrations for the next patient model.

Now if we take a look at the role of therapeutic drug monitoring -- I'm going to take the arrow out in just a second -- we know from our own experience of treating patients with tuberculosis that not everyone is a rapid responder to treatment, and these slow responses to treatment are common. And I'll show you that the CDC tracks this on their database. So, while many of these slow responses are, indeed, due to other things like treatment interruptions -- those can include adverse drug reactions or patients leaving the program, et cetera -- in our experience a substantial portion of these are due to poor drug absorption. So let's take a look at the CDC slide.

You can see that back in 1993, when the second surge of tuberculosis was peaking, only about 60-65% of the patients were on complete DOT, and that number has moved up dramatically, so over 80%, maybe 85% of the patients are on DOT, and I'll be showing on this slide, some of the results of that. You get a lot more people completing treatment. So back in 1993, only, you know, 60% of the patients actually completed treatment in a year or less, and now, with a lot more DOTs be used, you're seeing a lot more patients completing treatment in a year or less. But, still, it's only 80 to 85% of them, with about 90% of the patients completing treatment at some point beyond one year.

But, remember, this is supposed to be a six-month treatment regimen, and those original papers that I was mentioning, where we said they were 95%-plus effective, those per-protocol results, all of the people published in those results completed it in roughly a six-month period. So looking back at the papers and seeing what actually happens in the clinic, you can see that those two are not exactly the same.

So, remember, it's supposed to be a six-month short-course therapy. If it takes 12 to 18 months, then I don't think anybody would really call that short course, and you're paying for that patient that whole time. So if it takes 18 months to get them to complete the six-month treatment, it's almost like treating three patients. And, unfortunately, you're going to be paying for that as you're treating your patient over the longer period of time. So the longer they're on drugs, the more opportunity for adverse reaction or for

them to abscond from treatment. So, in theory, there's no difference between theory and practice. But in practice there is.

So I'm going to talk about some real basic concepts as far as the antibiotics, and you can argue this for any number of different classes of drugs, but today we're going to talk about antibiotics, and basically for every drug with a proven mechanism of action, this involves a drug entering an organism, binding to a target, and producing an inhibitory or lethal effect. This nice little picture over here, which is actually water molecules. You can see the oxygen in red and the hydrogen in white. But it just focuses our attention on the fact that this is a chemical reaction that has to take place, and it might be a reversible reaction or it might be covalent bonding. So, for example, when Rifampin binds to its target, it essentially stays on there forever and that target, intents and purposes, destroyed, and protein synthesis comes to a screeching halt.

But Rifampin, as I'm giving this example, is only about a little over 800 mass units or "Daltons," and if you consider a protein, that protein might be over 40,000 mass units, and cells are made of thousands of proteins. So from the standpoint of one molecule of Rifampin, a cell is a giant place, and your body is made up of millions of cells. So when we give that dose of 600 milligrams of Rifampin and how many molecules that actually counts up to be, they're being put into the body with no honing device. They're going to distribute all over the body and, hopefully, some of them, just by random chance, are going to bump into mycobacterium, enter that cell, and bind to the target inside a cavernous space from the standpoint of that molecule.

So you can almost think of this as kind of a semi-miraculous event, that the Rifampin actually gets to where it's going and does what it's supposed to do, and that's true for all of these drugs. The point, again, is that it has to be this actual physical binding of the drug. And if that doesn't happen, it's not going to work.

So if we look at the next slide, for every drug that we give orally or by some other route, typically by IM or IV injection, the only way for the drug to reach the bug or organism is through the bloodstream. In the graphic here the famous antecubital fossa, which is our usual sampling point right there. That's how we get the blood samples to see what's going on in our patient, for most patients. So if it ain't in the blood it ain't in the bug, and, therefore, pharmacokinetics matters. And you can see in this example that these organisms are having a happy time because you missed. So these mycobacteria are my concept of that. These guys are dissing you, so let's teach them a lesson.

This is just one example. And as I mentioned earlier, while there's not this prospective randomized study of therapeutic drug monitoring, there are accumulating data to show relationships between concentrations and outcomes, even if the study, per se, was not a therapeutic drug monitoring study. So this is from U.S. Public Health Service TB Trial 23, and this is the pharmacokinetics sub-study, and this was published back in 2005. And there was an association of lower Rifabutin concentrations, seen here, as an area under the curve or AUC over 24 hours. These guys with the lower AUCs, compared to these guys with the higher AUCs, had a 23-times higher likelihood of failing or relapsing with ARR, or Acquired Rifamycin Resistance. And as you know, if you lose the Rifamycin, it's just about as bad as NDR TB. So these guys did not have Rifamycin resistance going into the study, but because of poor exposure to Rifabutin, we had enough there to select out for resistance but not enough there to cure the patients. So I'm going to spend a moment on this slide so you kind of get it.

Most of the people had plenty of Rifabutin in their blood, and that's this blue box here. And then the outliers, with the higher concentrations and the lower concentrations, also were cured in this particular study. These guys, the bulk of them, had very low Rifabutin concentrations, as shown in the red, and these are the guys who had acquired Rifamycin resistance. Now one guy absorbed quite a bit of Rifabutin, now there might have been other things he malabsorbed, but the Rifabutin was in there in adequate concentrations compared to, say, these guys over here, yet, he did not have a good outcome. And that comes back, and we'll stress this later, when you're giving these drugs you have probabilities of outcomes. There's no written guarantees.

But, again, the point here is that the odds ratio for acquired Rifamycin resistance was really driven by the fact that these guys had low Rifabutin and not driven by the fact they had low CD4 counts. Yes, they had low CD4 counts, but the odds ratio based on just that parameter was only 1.04, and it was 1.00, with that zero effect. So the moral of the story is when you take your shots don't miss. See, the bug isn't laughing anymore.

Now, as you know, we have limited choices for the treatment of tuberculosis, and here is our fan club, if you will. We have para-aminosalicylic acid, Capreomycin, Cycloserine, Ethionamide, Ethambutol, Isoniazid, Pyrazinamide and either Rifampin or Rifapentine, also known as Cyclopentol Rifampin; Streptomycin, and then new improved Bedaquiline. We also use fluoroquinolones, such as Moxifloxacin and Levofloxacin Linezolid. And there are other unapproved drugs that, from time to time, we use. But keep in mind that we're going to use roughly four of these at a time. So you really get two good shots at somebody, and the second one is not nearly as good as the first shot, so you want to make sure, especially while they are on the first blind drugs, that you maximize your effect with those first-line drugs.

For you Grateful Dead fans out there, you remember this tune, Casey Jones, trouble ahead, trouble behind. Malabsorption or lack of blood flow to the sight of infection that you might see, say, with pleural infection or especially in old TB, where you have a calcified rind along the lung, you get reinfection or reactivation in there, there are certain situations where you have a fortress around the mycobacteria, and the drugs, even though they might be in the blood, are not getting into that particular fortress. Happily, that's atypical. But malabsorption is not quite so atypical.

The failure to identify and correct the problem reduces the choices that you have at your disposal. You start losing drugs to acquired resistance, and while it's not entirely common, it certainly does occur. And, of course, as also mentioned previously, while this patient goes from six months to nine months to twelve months on therapy, they're still under your care, they're still uncured, and you're still paying for. So, in the case of Casey Jones, he'd better watch his speed, and our equivalent is to watch the serum concentrations.

Now thinking about, you know, the case with Casey Jones and speed, you know, just about everybody on the call here drives a car, and I would hazard to guess that while you're driving the car, probably every minute or so you look at your speedometer, and there's a reason for that. You don't want to be going so fast that you get pulled over or you're at risk of an accident, and you don't want to be so slow that you have traffic piling up behind you or it takes forever you to get to wherever you're going. Although, I guess throughout the Southeast that is the case today, and you're not going anywhere.

Anyway, the analogous situation is when you start a patient on treatment, you give him the standard doses, you really don't know how much they're absorbing and if they're absorbing the normal amount or if they're going really slow or the equivalent slow and have low drug concentrations, and this is a way to demystify that part of you therapy.

Also, when you think about it, unless you're giving surgery to that patient, then you're completely relying on those drugs. They're the only show in town. And, again, you probably want to optimize those drugs. So the standard approach is to continue the same treatment and don't look for trouble. And, you know, over time, as David was mentioning, therapeutic drug monitoring has been, I guess, quote, unquote, a controversy. I personally don't see it as controversial. I think it's just like any other clinical test that you might choose to do, whether it’s a chem panel or CBC or CAT Scan or MRI. You may not always do CAT Scans or MRIs on your TB patients, but there's going to be selected patients where you're going to say, "I need to know more of what's going on in there so I can make a good decision." And in those situations you go ahead and get the test. And this is analogous to that kind of situation.

So talking about pharmacokinetics, we're going to get into a little bit more of the detail. It's the study of the movement of drugs through the body, and most commonly we're talking about serum concentrations. You can order CSF concentrations of the drug or pleural fluid or other specific fluids, but, generally, we're talking about the serum concentrations. And in the lower right we have this little graphic here, and this is sort of the classic description of pharmacokinetics. There's absorption and there's distribution. There's

metabolism and there's excretion, or sometimes listed as elimination. So ADME, you may see that from time to time in different papers, and that's exactly what they're talking about. It's the course of the drug through the body and, eventually, out of the body.

So applied pharmacokinetics is applying that general concept, which is often used in drug trials, such as phase-one and phase-two studies, and using it on a patient-by-patient basis in the clinic. You can see this guy here, he is now optimizing his dose, and he's taking a whopping dose of these drugs. So therapeutic drug monitoring is finding out just how many tablets ought to be on that spoonful for this particular guy, and every other patient after him. It aims to promote optimum drug treatment. And we can discuss what optimum might mean.

But, in general, you're the one that's going to be defining that for your patient, because you know that patient better than anybody else. You know if they're particularly ill, if they're particularly close to a bad event, including death, and the sicker they are and the more deathly ill they are, the harder you're going to push the drugs in all probability, and the more tolerant you might be of toxicities, because the alternative is going to be grave for that patient. So you're going to try to optimize the drug treatment by maintaining serum concentrations within the normal range, and that's just what typically would happen if you give humans these drugs. Or, preferably, a therapeutic range.

Now, unfortunately, when the TB drugs were introduced, most of them were just given as a single dose in all the different studies and it wasn't dose ranging in people. Some of that was done in vitro, and some of that was done in animals. But when it got to people, they pretty much settled on a dose and gave it. If these drugs were being developed today that probably would not be the case. And, in fact, we are conducting high-dose Rifampin, high-dose Rifapentine, and high dose Levofloxacin studies so see if there is a better dose than the standard dose used currently. And we'll come back to this graphic down here in the right-hand corner, but these are some of the terms that you're going to see, whether it's the Cmax, or maximum concentration; the AUC, or area under concentration time curve; Tmax, or the time that Cmax occurs; and therapeutic range. But we'll come back to that graphic in a few slides again.

So TDM, or therapeutic drug monitoring, is most useful when there is a direct relationship between serum concentrations and therapeutic response, and when serum concentrations serve as a surrogate for drug concentrations at the sight of action. So in TB we're often looking at the lungs. And while it is true that you can do lung biopsies, in general, that's not what we try to do in TB patients. Because if you thought about it, you would be introducing through normal tissue the needle into infected tissue, such as a TB cavity, and you would have the risk of spilling that cavity contents through the hole that you just created by boring in with your needle. So, for the most part, we don't do that. For the most part, we're going to have to make certain assumptions.

Now there's some really cool work being done by Cliff Barry and Vern eke Darter [ph] looking at PET Scans and seeing where exactly the drugs go in tuberculous lesions, and that's very interesting stuff. But I think the routine clinical employment of PET Scans is even further down the road than routine use of therapeutic drug monitoring. So I just mentioned that because it's very interesting work, but I don't think in the clinic you're going to be using that in the near future.

So TDM is most important when there is a narrow range of concentrations that are either effective or safe. And what we'll see on an upcoming slide is these don't necessarily track together. There are concentrations that produce efficacy and there are concentrations that may produce toxicity, and those done don't have to be the same in all -- those curves, in other words, they can be entirely different -- and when toxicity of lack of effectiveness puts the patient at great risk. And you can clearly say that for TB, and likewise, as you can see, for fungal infections, and you can say for HIV also, that lack of effectiveness of the drugs definitely puts the patient at risk. And we can actually do something about that. That's why I'm into pharmacokinetics.

So you might be saying, "Well that's great, Chuck, because you have pharmacokinetics in your job description and you went to school and took classes on that. But, you know, when I went to school for nursing or medical school there was very little discussion about pharmacokinetics, and, frankly, I'm just

pretty darn uncomfortable with it." And they may be exactly what you're thinking. So, happily, there are any number of decent books out there. This is simply one of them. There's another book by Winter, which is equally "spiftastic" as this one. And I have no stock in this book. I have no chapters in this book. It just happens to be a good book. And you can get this for laying down, maybe, I don't know, 70 bucks I think is the current cost of this. And you can go to the ASHP webpage, and there's probably others. You could just Google search and you will find this pretty readily.

And so given the fact that you guys are going to be using drugs to treat not only TB but if your clinic also sees STD or HIV or any number of other disease states, you have to use drugs to treat those. And if you're not fully comfortable with what's happening to drugs when we give them to people and you want to learn more about pharmacokinetics, this would be an excellent resource for your clinic or for your own personal bookshelf so that you can, at your leisure read about it. Turns out that this particular book, I really like the format, because the chapters are about three pages long and it's a very large font, so you don't even have to break out your glasses. And it's even got pictures in it, so what could be better than that.

Now as mentioned earlier, when I was talking about Tawanda Gumbo's paper at Bayesian kinetics and stuff like that, well Roger Jelliffe, who is the friend and mentor and colleague at University of Southern California, he had written this really, really nice paperback in 2000 in the "Journal of Therapeutic Drug Monitoring," -- there's a nice picture of Roger -- and it's entitled "Goal-Oriented Model-Based Drug Regimens: Setting Individualized Goals for Each Patient." And I would strongly recommend that you pull this paper and take a good look at it, because he lays out in four-part harmony all the kinds of things you might consider as you try to manage patients with, really, any drug.

Now his key points are that therapeutic concentrations vary by patient. In other words, shockingly, humans are outbred, and there's a lot of different genetic patterns across the population, and you can have people that are fast absorbers, slow absorbers, fast metabolizers, poor metabolizers, and all of these different variations, and. therefore, it's unlikely will that you're going to get the same concentrations in patient to patient. It's unlikely, in the case of TB, that every single isolate is identical with the exact same MIC, and, therefore, these things are going to vary from patient to patient.

So what he suggests is, rather than picking a dose per se, you pick a desired serum concentration, and, as mentioned just a few slides back, if you had a really, really sick patient you might pick higher concentrations to aim for. And then once you've pick the concentrations that you want, then you achieve these concentrations with the greatest precision possible. And that's where the therapeutic drug monitoring comes in. In other words, if you're relying on drugs to cure your patient, you may as well give the right amount of drug to each patient.

So this is a slide that really captures pharmacology just in one simple picture, and the thing I really will stress is that Y axis is probability. There are no guarantees if you have probability. So the yellow curve you can see as the concentration increases along the X axis the probability of the response approaches but rarely does it reach 100%. Usually somebody who doesn't respond for some reason. But you also see in this example that as you get above 30 micrograms per mL, the probability of toxicity goes up. Now we might say a priori, that we want to have at least a 25% probability of the response. And if we set that as our minimum standard right here at 25%, that's we don't want to go anywhere below that, then we would need at least ten micrograms per mL in if blood to achieve that. And if we did that, we'd have pretty low probability of toxicity. Now I might have oriented these red lines vertically at each concentration, and I can still make the point that way, but here I'm focusing on the probability of the outcome in this hypothetical example.

Now if we said, well I don't want to be below a 50% probability of target attainment, and you'll see that in the literature, then you'd need at least 15 micrograms per mil to get to that kind of response. Now you can also see out here, if you're concentrations are up around 38 micrograms per mL, you have a 50% probability of toxicity, but the difference between where it intersects the response curve and where it intersects the toxicity curve is sort of the therapeutic window that you can work within. Another way of looking at it if these lines were vertical, I had a vertical line at 20 and a vertical line at 30, then you can

see you have, at 20, this probability, and at 30, this probability of desired response, and in both cases, a pretty low probability of toxicity. But if I kept pushing, for this particular drug, the risk of toxicity or probability goes up.

Not every TB drug has concentration-related toxicity. Some classic examples include Ethambutol, and while the data are pretty thin, it appears to be the case for Cycloserine as well. But for other cases like Isoniazid and Rifampin, the bulk of the data suggests that those are idiosyncratic reactions. Now there may be some other data here and there that says, well, maybe it is related to concentration, but I would say the bulk of it at this point would suggest that it is not concentration.

So if you start looking at pharmacokinetics, that's the PK, and pharmacodynamics, that's the PD, papers in the literature, and there are plenty of them out there, particularly in the journal "Antimicrobial Agents for Chemotherapy" or the "Journal of Antimicrobial Chemotherapy," both of those tend to have these kinds of papers, you're going to see these different kinds of parameters. So the Cmax is otherwise known as the peak concentration, and the MIC is the minimal inhibitory concentration. And the ratio of those is one of the so-called "drivers of effect." We can look at the time that the drug remains above the MIC and I could actually put a slash in here, AUC divided by MIC, as another common parameter that's used for looking at how drugs work and how we will want to dose them.

So, giving this particular example, and we've got some exclamation points in there, so somehow this slide didn't translate happily into this media, but just avoid or ignore the exclamation points, unless you're really, really enthusiastic about pharmacokinetics. Anyway, in this example we have a Cmax of about nine micrograms per mL. We have a MIC of around three micrograms per mL, so the ratio is three. And for certain drugs, like the Aminoglycoside, Streptomycin, Amikacin, and for the Fluoroquinolones, Levofloxacin and Moxifloxacin, and for the Rifamycins, Rifamycin and Rifapentine, we want to maximize the Cmax, and we also want to maximize the AUC, relative to this minimal inhibitory concentration.

For other drugs, we might want the drug to stay above that MIC for just as long as possible. And for some drugs, say, like Cycloserine, we might want to give that twice daily, if that's possible, so that the concentrations are always above the MIC. We may want to do with that para-aminosalicylic acid also.

So as noted earlier, this was down in the corner of one of the slides up above, in here we have a better look at it. So on the Y axis we have concentration, on the X axis we have time, and this a concentration versus time curve, just exactly what the labels tell you, concentration versus time. And if we shade in this area under the curve, we have that area under the curve, or AUC. And you will see that a lot in the literature, but this what it's talking about, that whole area. And then, of course, can divide that numerical representation by another number, such as an MIC, or minimal inhibitory concentration.

Now you can also look at the peak concentration. And in this particular example, MEC is usually the minimal effective concentration, and the MTC would be the minimal toxic concentration, noting, of course, that not all drugs showing a toxic concentration anywhere near the doses that we're used. But in this particular example, this would be the therapeutic range. You've got enough to be effective above that minimal and not so much as you're going to be toxic. So these are the kinds of terms that you're going to see in the literature. And more and more you're seeing these terms in studies for tuberculosis treatments.

Now this is a really nice study that was done by Jayaram, et al, at AstraZeneca in Bangalore, India. It was published back in 2003. I had the pleasure of talking with some of these guys at a Gordon conference about a year, even less, before that. And what you see is, as you increase the concentration, in this case represented by the area under the curve of Rifampin, divided by its MIC for the test isolate in this mouse study, and this was a six-day acute infection mouse file. As you push that concentration higher and higher, you get more and more killing of the mycobacteria. In fact, they found the toxicity too high in any concentration higher than this. So the mice were still having more and more effect even at these really, really high doses, which were over 100 milligrams per kilogram, and the efficacy was still increasing at the higher doses. But for toxicity reasons in the mouse model, and well over 160 milligrams per kilograms, they had to stop pushing the doses higher.

But the point is, our usual dose is right here on the flat part of the curve. So as Denny Michison [ph] once put it in a conversation that I had with him, Rifampin at 600 milligrams is the minimally-effective dose of Rifampin. And while that's fine, we may want to try something above the minimally-effective dose. So the main reasons that we don't see clear dose response in the clinics are the following: not everyone got -- or I should say not every study used a dose escalation protocol, so the main reasons would include everyone in most of the studies got the same dose. So there's not that much variability of the achieved concentrations for us to pick apart what would be the best dose. Everybody got the same dose, so we can't really say that from a lot of studies, and the doses are at the low end of the dose response curve.

So let's go back to what we were just looking at. If we look at any number of studies, including some that we participated in from Batswana, the PZA concentrations had an association with outcome, but the Rifampin concentrations did not. And part of it is that all the concentrations were in this flat part of the dose response curve. There weren't enough high concentrations to really show the relationship in that population. And if we want to see that, we're going to have to give higher doses.

I am very happy to report that study 29X, which was conducted through the CDC TBTC has looked at much higher doses of Rifapentine compared to the originally approved doses from 1998, giving it daily and in doses more than 600 milligrams, and they're starting to see this kind of dose response curve. There's another couple of studies with Rifampin, one with Martin Boeree and colleagues, studying in South Africa, another that I'm participating in with Gerry Davies and Carole Mitnick in Peru, looking at higher doses of Rifampin, and we're starting to see this kind of effect, especially in Martin's data, some of which was presented at last year's Croi meeting, C-r-o-i meeting.

So now we're going to get to some of the real nuts and bolts about doing this. So let's say that you're modestly convinced that you've decided that you want to check the serum concentrations in one of your patients to see what's going on. Well how would you go about doing that and why? Well we typically suggest a two-hour and a six-hour post-dose concentration after a timed observed dose. So it is important that the events all are reported, and I'll show you how that's done in a moment. But nevertheless, for the most part, if you get two sample that is are separated by about four hours, and for most of the drugs it's going to be at two and six hours, you have a really good opportunity to capture the Cmax, even if something unusual is happening in that patient's gastrointestinal tract when there is delayed absorption.

So here is an example of Rifampin from some different studies that we participated in. So in the light blue this is what happened in healthy volunteers, and they had a lot of samples. They had 16 samples over 24 hours, not something that you're going to do in the clinic. Then, in the dark blue, we have TB patients in an NIH-funded studies and they had a series of studies, including the ones shown on the screen. And they ranged anywhere from, I think, five to seven samples, if I remember correctly. And then another group only had two samples, and you can see that the purple line and the blue line are pretty similar. That tells you that the two-hour and the six-hour samples, which was the only samples collected in purple, gave you roughly the same information as the dark blue.

And then in a separate study, this is an ACTG, or AIDS Clinical Trial Group study; these are all AIDS patients with TB, you can see that their malabsorbing the Rifampin. It's getting in very slowly, and it's getting in at about half the concentrations that we're seen in healthy volunteer. We just discussed the fact that Rifampin has profound concentration-dependent activity, so you would expect that as these concentrations drop, the activity drops with it.

Now let's look at a Ethambutol, same kind of graphic, same kind of colors, light blue, healthy volunteers. We've got in the extensively sampled TB patients -- let's see if I can go back up. It seems to have flipped forward magically all by itself there. Well, anyway, what you saw --

I'll do that for you. Just a moment.

Okay. One more back. There we go. Thank you very much. Well I guess this slide is so important it just wants to be shown. All right, so as this slide is settling in and Donna is fixing the equipment, what we see, again, is that the AIDS patients, in the dark green, you know, have less than the concentrations seen in

the non-AIDS/TB patients, who are in the purple, and they're in the dark blue. So, again, some of these patients malabsorb, and in those patients more likely to be seen in your AIDS population. I'm going to try to advance the slide here, and hopefully we get back to full screen. Are we able to get back to a full-screen display, Donna? Well while she's working on it I'm just going to proceed.

So, in this example, going back to Rifampin, we see the three basic patterns that you're going to see if you get a pair of samples. The yellow is what you want to see, a normal peak around two hours post-dose, and concentrations falling off after that. So that would be a very typical pattern, and in this case, it would be within the normal range -- thank you very much -- for Rifampin. So 12 micrograms per mil would be a nice peak for 600-milligram dose.

Sometimes you see this pattern in the blue, and if you only got the two-hour sample, you might think that you have to increase the dose. But if you have a two- and a six-hour you can see that in this patient, let's just say they're a diabetic patient and then you have some level of gastroparesis, the drug got in there, it just took a long time. Now you also can see this pattern if you give these first-line drugs on a full stomach. So you don't want to do that, generally speaking. Nevertheless, you can have delayed absorption, as shown in the blue, and then shown in red, that's malabsorption, and you really don't want that. And, again, because Rifampin has concentration-dependent killing, you're going to want increase this dose because these concentrations are starting to push downside towards the MIC.

And the other important point, which I'm not going to spend a lot of time on today, the MIC is a protein-free measurement, so a lot of these drugs have some level of protein binding in the blood, and the part of the drug dose that gets into the blood and binds to albumin or alpha one or acid nucleic protein, that's a depot, but it's not available to interact with the organism, so it's only the free drug. And in the case of Rifampin, about 85% of it is protein bound. So this number, which is representing the total drug concentration, really overstating how much free drug you have to treat the infection. Okay, so if you don't know where you're going, you might wind up someplace else.

If you were to order tests from our laboratory, and there's a number of laboratories around the country where you could get these tests. You may want to find out where it is that you can send your samples to and once you've determined that, you may want to find out what methodology is being used. I would recommend chromatographic methods such as HPLC or GC, which is gas chromatography. I would not recommend bioassays. Bioassays are fine if you're only treating the patient with one drug. But if you're treating them with multiple drugs, there is, depending on the bioassay, a possibility that all of the drugs are going to affect that result and give you a falsely elevated value. So that's something to consider. If you've, in the past, measured serum concentrations and you're not sure what lab tested it, the College of American Pathologists requires that the report states at the bottom the laboratory that actually did the testing, so you can still find that out. Going forward, you can choose whatever laboratory you're comfortable working with to decide how you want to do this.

If you look at our particular order form, these are the tests that we currently offer, and this an excerpt from the overall order form, and you can find that on our webpage. So here you have some of the HIV drugs, Amprenavir and Atazanavir. You have some drugs like Azithromycin, that are used commonly for non-tuberculous mycobacterial infections. We have some antifungal drugs, including Itraconazole and Voriconazole, and then we have the TB drugs, including Isoniazid and Rifabutin. Now you might see right next to it that there are recommendations for the sampling time. And as mentioned, usually it's two and six hours.

And if you look over here, you see Rifabutin. Rifabutin is just more slowly absorbed than Rifampin and Isoniazid. So for Rifabutin it's better to sample as three and seven times. So does that mean you have to poke your patient four times? No, it does not. I would recommend that if you have a patient who is on Rifabutin and say, Ethambutol, PZA, and Isoniazid, just give the Rifabutin one hour earlier, then give the INH, PZA, Ethambutol, and then two and six hours after the latter three drugs, you can collect samples, which will turn out to be three and seven. And you only have to do two venipunctures instead of four venipunctures, and now they'll appreciate that.

Other parts of the order form, I just stress that it's really important to fill these out. We want to know as much as you can tell us, reasonably, about your patient in these nice little boxes, and, importantly, we need to know the drug that you want us to test, the dose, the number of doses per week, because as we know, there are some three times a week regimens and the doses are different for some of those two- and three-times-weekly regimens, such as the doses of PZA and the doses of Isoniazid. We want to know the date and time of the last dose and the date and time of the blood draw. If you don't give us that information, we can still give you a result, but we really cannot interpret it, because we have no idea when it was taken in relation to the dose.

The other thing you can tell us is other drugs the patient is on. They might clue us in. So if you listed insulin, well, chances are that patient is a diabetic patient. Or if you listed Darunavir and Ritonavir, then chances are that is an HIV-positive patient, and that can help us interpret the results and tell you a little bit more about what you're seeing with your result and what you might want to do going forward. Not every laboratory is going to provide that kind of interpretation, but I think, from our standpoint, we consider that the most important part, not just the number, but what it is you're going to do with that number next.

So providing the detailed information allows us to provide you a detailed interpretation, and that detailed interpretation allows you to select the optimal dose, as already described, just as soon as you possibly can. Because the sooner you get the patient on the right dose the sooner the patient is going to start responding.

Now what do I mean by a detailed report? And these are excerpts from a hypothetical case of the patient named Joe Dirt. And, no, we never had a patient named Joe Dirt, although I think there's a movie by that name. And so this is not a HIPAA violation. I just want to be on the record showing that. And I'm making a composite of two different types. So in this example, let's just say it was past week or so, and we would have our own tracking number, the two-hour sample time point had two 2.01 microgram per mil. Well if you only got the two-hour sample, you might be thinking that you need to increase the dose. In case, the patient had delayed absorption, and you can see the six-hour values coming in at much higher than the first sample, and the second sample is in the range for daily dosing, but, unfortunately, this patient is on thrice weekly 900-milligram dosing. So even though this value is much higher, it's still low for the range for that drug.

Now, also from our laboratory, you'd get something that would look just like this. You'd have the numbers typed in up here, and you'd have some general description, and let's just walk through that for a moment. It says the normal range of Isoniazid is three to five micrograms per mil, two hours after dose, and then it goes on to say what the range is if you're giving the intermittent 900 milligrams. Well that range goes up proportionately from 9 to 15 micrograms. It also says that, you know, INH has concentration-related activity, and in this case it's AUC to MIC, so you're probably going to want to optimize that. Then it talks about specific patients. If they have hepatic dysfunction or if they have renal dysfunction, there's some description of what you might want to think about in those kinds of patients. And finally, you'll get additional comments, and let's go to the next slide so we can blow that up a little bit.

So what we do in our laboratory is we provide additional comments. If you happen to get samples from our lab, but they go through an intermediary lab, then they may or may not be forwarding this information to you. So if you're going through a hospital information system, they might only post the --

Hey, Chuck, we're no longer hearing you. Dave, do you want to get on and maybe take a question or two until we figure out how --

Yeah, actually, I mean right now -- I meanwhile we're waiting for Chuck --

Hey, I'm back. I was unceremoniously dumped, and I'm going to blame David, whether it was his fault or not. Okay, so for whatever reason, the telecommunication system decided it had enough of my voice and it just shut me up. But just being the obstinate person that I am, I'm back.

So what we provide are additional comments, and in this particular case the details state that the patient displays delayed and incomplete INH absorption, and I give a particular dose recommendation. Given what we're seeing, you may want to try a 1500-milligram dose. Now you guys are in the hot seat. You're the ones that are seeing these patients, and you've got to decide, given everything else you know about the patient, whether this is something that you're comfortable doing or not. But based on just the pharmacokinetic piece of it, this would be the recommendation, and if you were to follow that recommendation, all other things being equal, you would likely achieve the concentrations that would be considered normal.

What kind of tubes do you want to use? Well these are the tubes you don't want to use. And you've probably seen these in your clinic. There are purple tops and there's gold tops. And you can see that, depending on the type of tube, you lose drug. And then in an egregious example of Posaconazole, an antifungal, you lose about half of it if you use a gold top too. So the moral of the story is do not use purple tops, do not use gold tops, also known as zebra tops, SSTs, zero separator tubes, jell tubes, or any other term that might be used for these, if it's got that gooey stuff in it, that silicone stuff in it, don't use it. Use plain red-top tubes. If you have to, you can use green-top tubes for everything except the aminoglycosides, and that would be everything except Streptomycin, Kanamycin, Amikacin. We don't really know for sure if that's going to be a bad thing with Capreomycin as far as when you take the sample from the patient. In our testing it didn't look so bad, but that was an in vitro study.

So if you're giving the first-line drugs, I recommend giving these drugs on an empty stomach. So don't give high-fat meals, which would be pretty much any of these fast food chains. Don't do that. Well the patient might say "Well, you know, if I take these drugs on an empty stomach I really get nauseated, and I can't tolerate that." Well then give them a slight snack, and you give them a little bit, maybe not this much cereal, but a little bit of cereal, a cookie, graham cracker. But don't load them up with a high-fat meal, because that's going to substantially delay and somewhat reduce your concentrations.

Once you got them on a dosing schedule, with either snacks or an empty stomach, then you're free to test their pharmacokinetics, and that will represent what they usually do. If you're giving the second-line drugs, you may consider giving them with food like these yummy hot dogs or these yummy stews. And you can do that with Ethionamide, PAS, and Clofazimine, and that won't really adversely affect the concentrations, but it will affect Cycloserine concentrations, so I don't recommend a high-fat meal with Cycloserine.

For the most part, given the kind of data that we're going to acquire, unless we were to do that fancy-Bayesian in pharmacokinetics, which normally we're not able to do because we don't have enough information to do it, we're looking at the Cmax as a surrogate for the things like AUC. We can also get an idea of the trough or Cmin concentration, the time of the P concentration and the half-life, depending on the drug and the sampling that that’s done. Simple kinetics can be done with a calculator. We used to have drag races when I was on rounds back in Philadelphia at Presbyterian Hospital with Dr. Bonnie Vanider [ph]. We would have drag races to see who could do the pharmacokinetics for aminoglycosides faster. And we'd have to do it twice to make sure we did it right. So you can actually do this on a hand-held calculator. You can do it on a spread sheet, and I'll show you that on the next slide.

So this is a spreadsheet that I still use. It's very simple, and it's an over simplification of what happens in people, but it still gives us a pretty good idea of what's going on. So you can see, if you're capable of typing in four numbers, the two concentrations and the times that they occurred, on the spreadsheet that I've got set up, everything else just gets calculated. And so in this hypothetical example we have a half-life of about 2.7 hours, and that would be normal for an aminoglycoside, and you Cmax around 44 micrograms per mil, and the range that we shoot for is 35 to 45 micrograms per mil for a back calculated, like just exactly what I'm showing, the calculated concentration for Amikacin. But you see that the measured peak was only around 26.

And if you get it two hours after intravenous infusion or after an IM dose, the actual measured concentration is going to be lower than this calculated concentration, but the ranges are based on these calculated concentrations. And the advantage there is, it doesn't really matter if the samples are late. So let's say the patient went to radiology and they were late, and you got a three and a seven hour, it doesn't

really matter for an injectable drug. As long as the concentrations are above the limit of detection, we can still do these calculations.

So coming back to our probability curve, what we want to be doing is marching up the response curve while avoiding the toxicity curve, and all of this is a probability. But by choosing our concentrations judiciously we can enhance this probability and not really walk into this toxicity probability.

It is not possible to give drugs for the explicit purpose of avoiding toxicity. There is no guarantee about that. So if you really want a guarantee to avoid toxicity you have to avoid giving the drug. And, unfortunately, in the guidelines, if you start tracing the studies back to the original studies -- and I'll use the example of the BMRC studies -- you'll find that a lot of the doses in the current CDC guidelines are lower on a meg-per-keg basis than were done in those studies. So those studies, again, done in the '60s, '70s, and '80s, adult males weighed only 60 kilograms. And the meg-per-keg doses were considerably higher than what we commonly give.
So, for example, Pyrazinamide, in those studies that prove that Pyrazinamide should be a first-line drug, people got 35 milligrams per kilogram, but a common dose in the U.S. is only 20 or 25. And PZA definitely has concentration-dependent activity. So while I think the intentions were very good, with the hope that reducing the doses would keep the efficacy the same while reducing toxicity, I'm afraid it's the opposite. We don't change the toxicity for many of these because the toxicities within these ranges of doses are idiosyncratic, yet we do reduce the efficacy compared to the original clinical trial.

So the dosing of drugs, if you give the drug you must accept some probability of toxicity. There's just no way around it. The best way to avoid these toxicities is to give the most effective doses for the shortest time possible. You want to get in, you want to get out, and if you can get the patient off the drug in that six-month timeframe, then you spare them and everybody else the problems that come with extended treatment.

So the decision to use therapeutic drug monitoring is the same decision as should I check the CBC with this, or should I get a chem panel to look at liver function tests in this patient, or do I need to get a CAT Scan or MRI, or do I need to get any other laboratory or radiological test in my patient. None of these guarantees the treatment outcome, but all of these allows the clinician to make an informed decision, and that's why we would suggest therapeutic drug monitoring.

How much is the cost? Well at our shop if you're getting a pair of samples it's $70 per test. So that would be for two- and six-hour INH. If I could get someone to mute their phone, because we're getting a little bit of cross talk. Thank you very much.

So in this case, if you were to do two time points for all four drugs, it would set you back, for eight tests, about $560, and you still have the cost of packing it up and shipping it. Now let's just take a look at what I think are reasonable costs of treatment, some people have published it's $12,000, but let's just make the math easy and say six months is going to cost you $10,000. Well if the patient were to select out acquired Rifamycin resistant as one of the many problems you could face, well you've already laid out the original $10,000, and if we just treated, at the same cost, for another 18 months, then you'd add $30,000, all things being equal. So now you're out $40,000 in two years just in this particular kind of scenario.

It's certainly not the only scenario. I just use it to illustrate the fact that if treatment is extended, you have to pay for that, and that's a real cost. And now the $560, or however many tests you did -- it might be less than that if you did fewer tests -- that now doesn't look like such a bad deal if, in fact, it allowed you to avoid this. And I'm confident in the case of ARR associated with low Rifabutin concentrations that this is entirely avoidable.

So what is the role for therapeutic drug monitoring? It allows you to individualize therapy, and it allows you to optimize the pharmacodynamically-linked variable, whether that's the peak concentration or the AUC. It may, and I stress the "may," allow you to shorten treatment, and I think our colleagues up in Virginia actually have shown that in a number of patients and in their particular diabetic patients, and it may allow you to avoid concentration-related toxicity.

The part I really like is when you have patients who are on multiple different drugs. So it's not a shocking example to say you could have somebody who has HIV, and about 8% of TB cases in the United States also have HIV, and that patient may also be on anti-fungal prophylaxis or on treatment for an active fungal infection, and I've seen that multiple times. So now you have all kinds of drugs that are all interacting through the Cytochrome P450 system increasing and decreasing each other's concentration. And even though there are nice tables available from different web pages, including the CDC, that gives suggestions, almost all of those only deal with two drugs. So you might have Rifabutin and Ritonavir considered. But what if you add Voriconazole to that or if you add Rifamycin to that? Well there is no table that tells you what to do in that situation. So for complex multi-drug interactions, really, the therapeutic monitoring, I would argue, is your best way to handle that.

So what is the big idea anyway? In the end, knowing is better than guessing. If you want to read more about it, you can go to our SNTC webpage or you can go to our lab webpage. It looks like we've actually accidentally forwarded here one more slide. But I happen to know what's on that last slide, and I want to give a shout out to the guys in the lab who actually do the actual measurements of these concentrations, and that includes Vaneska Mayor, Behrang Mahjoub, TJ Zagurski, Kyung Mee Kim, and our office person, Roger Sedlacek. If you call our lab, you're almost certainly going to talk to Roger. Roger has been doing an excellent job with us. And with that, I'd say we can move to questions. I wish I had an answer to that, because I'm tired of answering that question. Thank you very much for your attention, and you can go to the questions.

Chuck, that was absolutely fantastic. Thank you so, so much. And, Chuck, you're burning up the computer lines. I think that's what happened. We've got so many questions, you know. But I really have to say, Chuck, while we're going to get right to the questions in a second, I think there's one burning question that needs to be answered. And, Donna, if you don't mind putting that up, I think we need to start off our question and answers this way, which is, you know, there's a big thing coming up on Sunday, Chuck. And, you know, I do believe, if I'm correct, that you are from -- what state again, prior to this.

Color auto.

Yeah. So I guess if you don't mind, I think the first polling question we have to have is, like, who do you guys want to win the Super Bowl, Denver or Seattle? So let's see how that happens, because I have my bookie on the line, so I need some help here. But, Chuck, while that's coming in, you know, we have so many questions coming in. And one of the questions keeps coming up, Chuck, I mean if you have to say, hey, look, the first question, Chuck, should we doing drug levels on everybody? And I know you're going to be a little bias on this. But what do you think, Chuck, is there any study that we do it on everybody or is there selected crew? I mean should we wait for people to fail, you know, which is one of the recommendations? And, Chuck, I mean in general, you know, do you think there's any evidence for routine -- and I know you gave some numbers on what it cost, but do you think there's evidence for routine, and if not, if I have to turn it the other way, who do you definitely think needs drug levels?

All right, so, no, there's no prospective randomized study that's going to tell us exactly who. And so there are a couple of ways of thinking about it. And it really it kind of comes down to the old Fram oil filter commercials, pay me now or pay me later, and I'm not talking about me, just paying in general, okay. So if you simply go and give the standardized doses, and if things aren't going swimmingly, you know, you keep getting sputum measurements to see what the smears and the cultures are doing, and you just keep pushing ahead, well, according to the CDC guidelines, if I remember correctly, if you get to the four-month point and the patient is still culture positive, which, of course, you're not going to know immediately, but that's a failure; right? So obviously you don't want to get to that point.

Now the old dogma was that, well patients who fail always fail with drug-susceptible TB. And we now, given all the MDR and, now, XDR around the world, that's not the case. So I would say the sicker the patient and the more difficulty they have, or the greater risk of a really bad outcome, then those are the patients you need to get it right and you need to get it right away. So I would not hesitate to do therapeutic drug monitoring to anybody that had to go to a unit or anybody who is coming in with, say, a

hemoptysis so they've got a major, major problem, and you may want to think about getting the doses right right away.

Other patients -- transplant patients, renal failure patients, aids patients -- I would have a very low threshold. And I can't point to a particular study that says, "Oh, you must." And, again, it's the kind of thing where you have to decide, as a clinician, is this information going to allow me to make a decision? I mean we don't want to just populate the chart of the patient. The numbers themselves are not going to do anybody any good. But if it allows you to say, "I'm giving the right dose," or, "No, I'm giving the wrong dose and I'm going to change it now," if it's going to let you make that decision on a timely basis, then I would suggest that you get the concentrations.

Yeah. And, Chuck, I mean let's go -- you know, I totally agree. I mean I think there's a lot of questions out there about it, so I'm just going to put them all together, you know. But I think you'd agree. You mentioned some of the classics. I mean some of the others, you know, when it comes to patients with liver disease, as you mentioned, patients, were seeing more and more on transplant patients, on other medications, where therapeutic drug levels, not just for the TB levels -- I think you'd agree -- but also for the other therapeutic drugs, the immunosuppressive drugs becomes huge, if I'm correct.

Yeah. So if you're giving calcium urine inhibitors or even Prednisone, then in those situations, Rifamycins are going to dramatically change that. And really common for these patients to be on fungal therapy, either prophylaxis or treatment of an active fungal infection, so they may well be receiving Itraconazole or Voriconazole, and those drugs are going to be cleared by enzymes that are going to be affected by the Rifamycins. And Rifabutin, unlike Rifampin, is partially cleared by Cytochrome P453-A4. It's partially-active metabolite is completely cleared by 3A4, so you have Voriconazole and some of these other drugs blocking the clearance of Rifabutin, so you have a two-way drug-drug interaction. We've actually published a case report on that, and people can look up on PubMed. They can put in my name and put in Voriconazole, and they should be able to find that paper in pharmacotherapy pretty quickly.

So when you get into those kinds of situations, you should, obviously, be thinking about measuring the Voriconazole concentrations or the calcium urine concentrations and making sure that their Cyclosporine or Sirolimus or whatever drug that they're getting is being dosed in such a way to put them in the normal range. And when you get into these situations with high-risk for drug-drug interactions, Rifabutin tends to be the preferred Rifamycin to use. But Rifabutin is not exactly the same creature as Rifampin. It has concentrated-relation toxicities, and it has two-way drug-drug interactions. So of all the different TB drugs, I'd say Rifabutin is the poster child for therapeutic drug monitoring, and it's been proven that if you get the dose wrong, you select out for Rifamycin resistance.

And, Chuck, I agree. I mean just a little -- to go over that a little. I mean first of all, if you ever want to make a cost analysis on transplantation, I think they're the ultimate. I mean in the sense that so much money has been invested in those organ transplants, and you have very little window of -- you know, you have a very small therapeutic window to, you know, that could endanger not only the patient with TB but also injection.

But when you think about it though, there's a lot a number of questions about it. You mentioned the Virginia, you know, data. And you now offer you mentioned you could prove it. I mean do you have any paper -- I know you said you put in your name and, you know, with can search the Internet for some of the data, but a lot of people worry about put your name in on their Google web browser.

Get the FBI showing up, yeah.

Right. Yeah, especially for us who work in the state, I mean it would be a problem. But I mean do you have any -- you know the Virginia -- do you want to talk a little bit about the Virginia experience?

Well I think for those who missed it the first time, Eric Hoff [ph] gave a great presentation on this very format, and I think his presentation is available on the SNTC webpage for those who missed it. But the bottom line is that they were seeing an increasing number of patients with diabetes, and, very frequently,

those patients had either delayed absorption and/or mall absorption, or usually both, or certainly malabsorption. And rather than waiting for those guys to kind of sputter along and not get better for a while, they've elected to, early in the course of treatment, test your diabetic patients for the therapeutic drug concentrations and then adjust the doses right from the start. And in their experience, by doing that, they've shortened the total duration of treatment.

And it was a great presentation, and, you know, forget the presentation, I think all of us who deal with diabetic patients, which I think we're seeing more and more, I mean we've all lived with this, and I totally agree, I mean, with Eric that, you know, it's definitely one of those findings that we used earlier -- I mean to do it earlier in. And Donna has been kind enough to put up the TB and diabetes webinar address up, as well as your paper, Chuck. And the reason they do that, again, is to protect web browsers and some the Spam they may get after that.

Well safety first.

Chuck, you know, what do you recommend? I was talking to you before this, and I see a number of questions. We get asked this all time. You know, you have a renal patient and, you know, how should you dose the meds, and is there a role for TBM, especially when you're using INH or Rifampin, which are plainly hepatically, you know, metabolized? But, you know, what's the role of TDM in patients on renal dialysis, and when do you recommend giving the meds, you know, and what doses, if you don't mind?

Some in the 2003 guidelines there actually is a segment on renal failure dosing of these drugs. And it's my favorite section in the guidelines because I wrote it. Just kidding. But I did write it; that part is true. But there's plenty of other sections that I like just as well.

Anyway, it is true that INH and Rifampin are largely cleared, but not 100% cleared hepatically. And in a acetylator Isoniazid, which, of course, just looking at the patient, you can't tell, they have a little bit more reliance on renal clearance of Isoniazid, and the metabolite goes out to the kidneys as well.

Now taking a step back, so now you have a patient who is on hemodialysis or perineal dialysis, or they're a candidate pretty soon for dialysis. Well how did they get that way, and in many cases they're also diabetics. So they have all the sequelae from long-term diabetes, including renal dysfunction, and they may have gastrointestinal dysfunction. So, often in these patients, they're not just patients who have poorly-functioning kidneys. They are patients who have multiple system problems. So I would have, personally, a very low threshold for checking those patients because they're also the kinds of patients that Eric was talking about, who are going to mal-absorb or have really delayed absorption of their TB drugs.

The other thing in those patients, is you may be using Ethambutol, and Ethambutol is one drug that you absolutely positively have to change the dose if the patient has renal dysfunction, because, as you know, you can have that drug accumulate, and that can lead to optic neuritis and that can, in turn, lead to blindness. Pyrazinamide, we hedge our bets. So in the guidelines we elected to change that to three times weekly dosing in hemodialysis patients, just like you would with Ethambutol, because we know that the metabolite, or metabolites, pleural, go out through the kidneys, and we don't know if it's the parent drug or the metabolites that cause the liver toxicity associated with PZA. There's just no data to speak to that. So hedging our bets, we decided that giving the intermitent daily dose, but intermittently for both Ethambutol and Pyrazinamide are reasonable thing in these patients who have end-stage renal disease.

So just to clarify, Chuck, what you're recommending -- usually what we do is we give the drugs right after dialysis. Actually, we usually, after dialysis, say to themselves to give the drugs. Usual patients get it three times a week, and you keep the three times a week dosing on the INH and Rifampin, but then use a daily dosing but three times a week on the Ethambutol and PZA. Is that what you recommend?

Yeah. I think you want to give them as much drug as you can, and there's been some nice papers by Wing Lee Yu [ph] and colleagues looking at the cost of going from seven days a week to five days a week to three days a week to two days a week. Now this was not specific to end-stage renal disease. But with each decrement in dosing you lose a little bit of efficacy. So I would suggest for these sicker patients that

if you can give it seven days a week, great. If you can't, five days a week for the INH and the Rifampin. And then you give the daily dose size, but give it intermittently three times a week after dialysis for the Ethambutol and the PZA. Now it's best to give it after the dialysis so you're not sucking the drug out as you're putting it in.

And the other thing is if you want to do DOT there's really no better place in a dialysis clinic. If you've got somebody's arteries hooked up to a bunch of tubing, they ain't going anywhere. So you have a little leverage to get them to take their drugs.

Well, and also, I think, from a TB perspective, I agree with you, Chuck. But the dialysis staff wants to make sure that person's getting their drugs, because as you know, all you need is a case of TB in a dialysis center. That's never a great contact investigation. And just real quickly just to, you know, kind of sew up this dialysis. You know, a question is coming in, "How long does it take for the blindness to occur?" And I think you'd agree, it's very hard to say that. But I mean, it is related to the level of the drug, which we do know, if I'm correct, with Ethambutol.

Yeah. In the studies that originally showed this, only a small number of them actually had serum concentrations. But they all had the constant theme of giving standard doses, daily doses to people who really should have been getting the drug intermittently. And we know from the kinetics of this drug, which are reasonably well studied, that the drug is going to accumulate. It has no where to go. I mean there is some portion of it that is metabolized, but a lot of it is through the kidneys, so the drug is going to build up. And, clearly, there have been multiple lawsuits over this, so I can guarantee you, if for no other reason than the medical legal reason, that you want to be very careful with Ethambutol in patients with renal dysfunction.

I totally agree. And just one other thing, you know, getting into the issue, you know, a lot -- personally you know, we talked about, you're right, I mean most people say about four months of culture positivity you'll failed. We, in general, as you know, it's been shown about anywhere from 75 to 85%, and it depends on what studies you're looking at and from what part of the world you're looking at, which we'll talk about in a minute, you know, are culture negative by two to three month, if not higher. So normally, in Florida, if we have somebody who's still culture positive on their two-month culture -- I want to emphasize when I say "two-month culture," remember that I'm talking about collective. Let's say the person had TB and was diagnosed on January 1st, and we're collecting that March 1st culture, and then, as we all know, on April 15th, you know, six weeks later, that's positive. If that person is still positive on the specimen that was collected at two months, we usually, then, start going for drug levels.. Is that something that, you know, you see as appropriate?

Well, yeah, if the patient is not getting better, then you have to go through the list of, okay, what could be going wrong. And first you have the wrong diagnosis, or they have two diagnoses and you're only treating one of them. So it is not impossible for someone to have TB and no cardia and TB and an NTM, or TB and a fungal infection, and they can both be in the lung, and they can even be in the same part of the lung. So you want to make sure that you've got the right diagnosis and there's not something else going on. Then there's non-compliance. Including occult non-compliance, and David and I have seen some really unique cases where patients go to the extremes not to take their drugs, even when they're in a TB hospital.

You know, I don't want to say nothing, Chuck, but that's HIPAA information about me and I don't think you'd be sharing that with my non-adherence.

Okay. Well I won't talk about your electroshock therapy. Okay. So as Andy Vernon succinctly put it a number of years ago, he goes, "Chuck, if these were $5 a test everyone would be doing it." And he's right. But unfortunately it's just not possible to do that. The equipment is very expensive, the consumable supplies are very expensive, and paying for the chemist is expensive. And they're really nice people, but they really don't work for free. They want to get paid, and I can't really blame them. And that's true of any laboratory worker. In our laboratory, or any other laboratory you might use, you'll find that there's a certain price range out there for these kinds of tests, and that's just not avoidable.

So the question it comes down -- and this hearkens back to some of the earlier questions -- do you test everybody and do you test them all at two weeks? But I don't think most TB control units around the country would feel comfortable doing that. They probably just don't have the budget for that. And that doesn't speak to whether it would be a terrible or not, just, you know, they would get concerned they were getting more concentrations than they might otherwise need. So you probably need to triage the patients, and so, again, you know, the sicker they come in, the more I would consider doing therapeutic drug monitoring. The more concurrent illnesses or the more complications they have the more I would consider doing therapeutic drug monitoring. If they're on Rifabutin I would do it and --

Whoever is on, can you make sure, please, you mute all lines, please. I'm sorry, Chuck. All right, let me ask -- you know, you one of the questions that we're really getting a lot it on, is how does different factors affect drug levels? Like how does age affect drug levels? Do you make any adjustments for that?

Well the most important adjustment would be in the pediatric population, and there's some really nice studies out of South Africa, where Peter Donnell [ph] and colleagues have clearly shown the dosage that we've been giving all these years, if you give the standard adult dosage to children, you under-dose them. And WHO hasn't updated their dosing guidelines. I imagine that's going to be incorporated into the new ATS, ITSA, CDC guidelines as well. So if you're treating a child, chances are they're going to have very good renal function and very good liver function, and they're going to blast through the drug. So, especially for our INH and Rifampin, the doses should be proportionally higher, pushing up towards the 20-milligram per kilogram dose for Rifampin, rather than the more common 10-milligram per kilogram dose for adults.

And the other end of the extreme, as people age, they lose some of their creatinine clearance or their renal function. So for the drug that is are renally clear, you need to keep that in mind. So if you have an 80-year-old patient who had TB a long time ago and now they've got a recurrence, and you, for whatever reason, have to use, say, an injectable drug or Cycloserine or Ethambutol, then you might consider still giving the same size dose, because you still have to get over that minimal inhibitory concentration, but at least for selected drugs with renal clearance, you may elect to give that selected drug only three times a week while you're giving the other drugs five times a week that are hepatically cleared.

You know, and so you're saying in children, and, you know, are we talking about infants? Are we talking about toddlers? What age groups are we talking about? And is there a difference between, like, in infants you should definitely be doing TDM, or in toddlers and children. Chuck, you know, how do you define it?

Well I think, first, people would be well served to look at new guidelines from the WHO, and also to look at the papers by Peter Donnell. I think happily, with children, they often have relatively paucibacillary disease, so they typically don't have large cavitary lesions with huge populations of drugs -- bugs rather. They often tolerate the drugs pretty well. So I would not say that the standard of practice is TDM in all children. I would not say that. Selected patients who are pediatric patients may need therapeutic drug monitoring because they fall into one of those complicated categories like a transplantation, or they're really not doing well clinically. But I think if you talk to experts on pediatric TB, including folks down the Houston, in including can have Jeff Stark, but certainly not limited only to Jeff, I think a lot of people would say, yeah, you probably need to give bigger doses than have historically been given in the United States, but you probably don't need to do TDM on all of these kids.

Right. And how about things like weight, Chuck? I mean we're getting questions does weight affect it, does ethnicity affect it?

Well weight, clearly, will affect your milligram-per-kilogram dose. And as I mentioned at the outset, the BMRC studies of INH and Rifampin and such, you know, the patients, the average weight was 60 kilograms. So if you gave 600 milligrams to a 60-kilogram adult male, that's ten megs per keg. If your patient 90 kilograms, you're not giving ten megs per keg. And the drug has concentration dependent killing.

Now the question comings up, should it be the total body weight or the ideal body weight? And the answer is nobody knows. There's been a little bit of work in the area, I think Tawanda Gumbo has done some work with Ethambutol on this topic. So if you have a morbidly obese patient -- and, you know, you just didn't see those before. I mean after all, TB is called consumption. People would come in and they were cachectic. Now we're not seeing exclusively that phenotype. We do see people who are morbidly obese and they have TB. So in that patient, I would probably start them on doses based on their ideal body weight, the standard formula, you know, 50-kilograms plus and all that stuff. You've seen all these formulas out there.

So I would start on the standard doses based on their ideal body weight. But early on, after a week or two, I would do therapeutic drug monitoring on those patients because almost for sure some of those drugs are going to be dramatically under-dosed, given the size of the patient. But I can't tell you beforehand which drugs and how low they're going to be.

Yeah, Chuck, I totally agree. I mean, as you know, over the years we've treated a number of patients who are, quote, unquote, morbidly obese, and it's like there's no way to predict the right dose, and the only way is TDM. And we really have utilized it a lot. You know, I guess, you know, the question comes down to, and one of the questions being asked here, I mean, you know, how many -- chuck we do a lot of, as you know, we do a lot of drug levels in Florida, and, you know. In general, I mean when we do drug levels on a large amount of our patients, the vast majority, I would say at least half of them, depends on the drugs, have lower than expected levels, you know. So I guess the first question is, Chuck, what does that mean clinically? I mean, you know, do you think that all these patients fail or do you think that the most of them fail -- I mean most of them do well but you just don't who is going to fail?

Well I would say there's an entire spectrum there. And we do, again, if you look at the BMRC papers and some of the U.S. Public Health Service papers, again, people were smaller and the people who ended up in the final analysis were per protocol. So they basically took all of their doses. Those people did really well. It was 95-plus percent long-term cure rate.

Now let's look at the clinical situation. First of all, in the U.S. we follow the patients through the completion of treatment, but we rarely follow them for relapse. We're making the assumption that the rates of relapses are pretty low. Now there may be exceptions where certain clinics do follow people long term, but at least in the slide sets that come out every year from the CDC, there's nothing in there about recurrence post-treatment. It's either you're cured or you failed at the end of treatment.

As far as, you know, are these going to fail? No, they're not. But this comes back to some of the things that Tawanda Gumbo talks about and I talk about in the papers that I put in the early slides, and then what Roger Jelliffe has spent his whole career looking at. So rather than picking a dose, you pick, a priori, what concentrations that you want to get. Now you may want to just assume that for some patients you're just going to give them the standard doses, you're going to just watch, and if they're doing great, well, then you've got your answer clinically. In other patients, you're not going to be comfortable doing that. And that really is a clinical call, based on the reasons already stated as far as comparing illnesses or acuteness of the illness.

But what I would suggest, if you're doing therapeutic drug monitoring, if you've already made that decision, then there's no reason to keep them at concentrations below what we consider to be normal, because for all of these first-line drugs we have a pretty good idea that AUC divided by MIC is the driver. And now we have some clinical data, particularly for the Rifamycins, that if you don't achieve those, you don't do as well. Or, on the other side, with increasing doses of Rifapentine and Rifampin, if you get higher concentrations the patients do even better. So there's enough pharmacodynamics data to suggest that these things are real. If you elect to do the TDM I would say, change the dose to achieve the concentrations that give you a higher probability of the response that you're looking to get.

Yeah, and, you know, one thing about this, because this always comes up, one thing I found to be -- I wonder how you feel, Chuck -- is that what's very interesting is that, you know, if you read the PDR, it will

tell you that if you raised Rifampin to above 600 milligrams a day, the Earth splits in half, the heavens come apart. And the one thing we find is that, you know, if you raise it, as long as the levels seem to be within that expected range, interestingly enough, it's very, very well tolerated. Do you agree with that?

Yes. And I think the reason that the package inserts are written the way they are is that the package insert is an agreement between the FDA and the drug company as to what type of discussions they're allowed to have with clinicians. It's basically a legal agreement. And unless the company has some real compelling reason, they're not going to revisit that with the FDA. And in the case of the TB drugs, with the exception of the Bedaquiline, they're all off patent, right, so they're all generics. So there's no incentive for anybody to go back and visit the package insert; right?

So, that said, you know, you have to make the call as a clinician, if you've got objective information that your patient has a third of what's considered the normal concentration, then, you know, all things being linear in that system, you're going to need to double or triple the dose to get them where they need to be. Now, in the recommendations that I give I rarely will recommend anything more than a doubling of the dose. But it is fairly common that people go from 300 to 600 milligrams daily, and that's what they need on INH to get them into the normal range. And it's not uncommon to go from 600 to either 900 or 1200 milligrams to get them into the normal range with Rifampin. My personal record is 2100 milligrams a day of Rifampin just to get them to eight micrograms per mil.

Yeah, and I would just back it up. Our record for INH is 1500 milligrams a day to get them into an expected range.

Yeah, so in that situation, the drug is going out in the stool.

Right. Well, you know, and here's the funny part about these cases – and, you know, I want to talk about specifics because there's a lot of questions. But there comes a point, I think you agree, Chuck, that you can't trust the GI tract after a while, and you come to those issues, you should you be switching to IV therapy, and I know that's another grand rounds altogether. But, you know, there are those cases where no matter how high you go, you're not getting anywhere, and then IV therapy becomes a real alternative. Do you agree?

Yes. And if you look at, you know, older textbooks for pneumonia, you know, a lot of -- you know, Harrison's older editions and that, you know, some of the original papers on antibiotics for pneumonia, the constant across those is that in an acute pneumonia patient you don't trust the GI tract. You give the drug intravenously. So now let's look at tuberculosis. It's a form of pneumonia in the majority of the patients that we're talking about. And if you have an acutely ill patient with a TB pneumonia, well, the same kinds of things are happening. And if we look at what happens in consumption, you know, you have cachexia, or whatever it is that's in their blood, you get a generalized derangement, including a generalized derangement of the GI tract. That's why they're losing so much weight. You know, they're not eating, and what they're eating they're not absorbing properly. So it's not limited to TB drugs as far as malabsorption in patients with really bad pneumonias. An acutely ill TB patient can be seen in the same light.

Right, I agree. And so let's get back to nuts and bolts, as we said. You know, there's lots of questions -- so let's see, Chuck, get back now to the low level. And I want to make a very important point you made, and that's -- again, I'm really biased but, you know, one of the keys that I think is a really unique aspect of your lab is that when you get the levels, you also give recommendations to how do adjust it, you know, that how much you feel -- you know, you give a true pharmacological recommendation, which is for a lot of us very, very helpful, especially to those that may not have a pharmacist at hand or a pharmacist that is, you know, familiar with TDM. We got a couple questions asking for programs, you know, which program would you use to calculate it? I mean, personally, as a clinician, I would just say I try not to. I leave that to my pharmacology colleagues. Do you agree?

Well, yeah, there's not a package out there that's ready-made for the TB drugs. The simplest thing is, you know, I use my own internal Bayesian program, you know. So Bayesian, you know, involves having a prior expectation, and then getting feedback, in this case serum concentrations, to see if what you

thought was going to happen is what happens, and it allows you to adjust. And basically that same kind of Bayesian approach is exactly what's used to fly airplanes. So you tell the airplane where you want to take your turn, and the computer keeps checking all the sensors around the aircraft to make sure that the plane is now moving in that direction. So that's the new fly-by-wire approach to avionics.

For purposes here, a lot of the times for oral drugs we're making certain assumptions about, you know, the rate and completeness of absorption. So, oftentimes, we don't really do sophisticated calculations, if you will, not that those would be a bad thing, but you'd really need what Tawanda Gumbo was talking about in his paper and what we've published in some of our population PK studies, you need a population model and a program that can handle that. And there's actually very few number of programs available that allow you to do that. For the most part, the drugs that you're probably going to want to calculate half-life on would be the aminoglycosides. And I just do that on a spreadsheet, but you can do it with a handheld calculator. And that book by DiPiro that I showed tells you exactly how to do it. So it's actually pretty easy. You can even do it with semi-log graph paper where the y-axis or the log scale axis is the concentration, and the x-axis or normal scale axis is time. And you can just, you know, put the dots on there and draw in the line. And from that you're off to the races.

You know, Chuck, it sounds easy to you, but when I use my crayons on it, I just can't draw in the lines, that's the problem. So, Chuck, now we increase the dose, right? And so I guess the next question comes down to once we increase the dose, Chuck, do you recommend repeating the drug levels, and if so, how soon?

Well, you know, that really depends on the clinical situation and the need to know and what it's going to allow you to do. So, you know, for the people that are extremely trusting, then they could accept the dose recommendation and make the change. For those people who are more like Ron Reagan, but, you know, they trust but verify, then in those situations you can go back and you really can do it on the very next dose. For most of these drugs, they're short half-life drugs. There are exceptions like Rifabutin. But for INH and Rifampin, in particular, there is no true steady state. They have very short half-life, and in 24 hours the drugs are gone.

So if today you gave 600 milligrams of Rifampin, and then you got your results back, and it said, oh, you need to go to 1200 milligrams Rifampin, and you did that. Now that the patient has already undergone the auto-induction weeks ago by now, then, you know, you could just go tomorrow if you wanted to, check the serum concentrations after the higher dose, because that dose is going to be independent of today's dose, because at 24 hours there's nothing left from today's dose.

And, you know, just, again, we got a couple questions about, you know, do you – you know, when you're doing those doses, you know, you're doing the two and the six, I mean, I guess the question that we're getting is do you really care about how many hours apart the two levels are drawn, especially if you're using the two-level kinetics? And the other thing is, does it have to be at least one half-life apart between the two levels?

It depends on what you want to do with the data. If you want to calculate half-life, and that would be the case for the aminoglycosides, a lot of times you want to know that information,then you don't want to be extrapolating through your half-life, you want to be interpolating. So what does that mean? So the space between the two dots should exceed the half-life of drug. Now the usual half-life, as I gave in that example of the spreadsheet was between two and three hours for streptomycin or Amikacin. So if you sample four hours apart you're going to be able to interpolate the half-life of that patient, and that's a good thing. If you had a three and a seven-hour – three and a seven-hour sampling you still can't really calculate the Rifabutin half-life. The data don't really speak to that Rifabutin as complicated to compartmental kinetics, not going to spend time on that.

But the bottom line is that two and six hours, even in a patient with gastroparesis, they're going to be absorbing by at least six hours, okay. Or if they took it with a meal, they'll still be absorbing it by six hours. After that, there's just not a lot to be gained other than really ticking off the patient if they're an outpatient by making it hang around later. Rifabutin is a different thing. I try to get seven hours if I could, right. Now,

let's say, well, in my clinic, you know, and patients aren't going to hang around and they're yelling at me, and it's like four-and-a-half or five hours after the dose, I'm just getting that second sample now, and to heck with it, you know, that's a clinical decision. Give you a chance to look at the late absorption if that's what's going on in your patient. But it's been our experience that two and six hours does a pretty good job at a number of different things, including picking up the late absorption. So in the situation where you can wait that long, I would recommend it.

All right, now, Chuck, here's the million-dollar question, and we always get asked, and I'm really interested in your perspective on this. Okay, we changed the doses, voila, we now have really, really good levels. Everything's within the expected range. But, unfortunately, it took four months for it to get into that expected range. Do you extend therapy? Is there any guidelines to how long to treat? You know, do you go beyond the six months? If so, how long? What do you use, Chuck?

Well, you know, I think it depends if you're living in a red state or a blue state. If you're conservative or you're liberal. So, you know, or maybe it's more of a Dirty Harry question, you know, do I feel lucky, right? So if you had extremely low concentrations of the drug, like trace, or really in the lower quarter of the range, I wouldn't even count that, personally. If they were halfway or three-quarters up the range, you know, they're probably going to be getting a lot of benefit from that, because there's going to be a range of concentrations that are effective.

The other thing is that in TB we almost always get susceptible or resistant. We don't have the MIC or minimal inhibitory concentration. And there's a range of MICs. So if you're lucky, then you got a patient with a really low MIC, and therefore you're really pounding that bug, even with modest serum concentrations. But if you're unlucky, then you got a real high into the range MIC and now you don't have much separation between the drug that you got into the patient and the amount that's needed to inhibit.

Now if we just take a quick step back, almost everybody uses susceptibility data. And the actual coin of the realm in susceptibility testing, unless you're doing genetic testing, is a concentration, a minimal inhibitory concentration. And the term is important. If you're below that, you're below the minimum needed to inhibit or kill the bug. So if you use susceptibility testing, you actually, unbeknownst to you, have already bought into the idea of concentrations. It just goes back that the assumption is if you just give the drug you'll have enough in their body. And I think there's plenty of data now to show that there are plenty of patients where that's just not true. And that's where therapeutic drug monitoring comes in.

Yeah, and I agree. I mean, I think you'd agree, there's no hard facts or -- these are great guidelines. And we do the same thing, that once we get the drug levels to an adequate range, depending on the clinical response, depending on how low it was, if we can keep that person above there for about four months, you know, we've done very well.

If I could jump in --

We're running out of time, but, boy, there's so many questions. I want to stop here and thank everybody for their questions. All these questions that are being asked are really compilations of, you know, a tremendous amounts of enthusiasm, and great questions. Sorry we're not going to be able to get to all of them, but I do believe that we'll be able to get some more of these questions answered later by e-mail, Chuck.

But, Chuck, one other question that, you know, I have for you, which is the famous question about, you know, what about TB drugs and illegal drugs? Is there an effect on drugs like alcohol and the drug levels, and do you think that's why patients tend to, A, potentially fail more, develop more issues with resistance, as well as liver issues?

Wow, man, well, I used to know the answer to that, but I forgot. So ethanol is a solvent, so ethanol is often used in pharmaceutical solutions that, you know, you get at your pharmacy that are called "elixirs" is the technical term. But, you know, a lot of cough syrup, that kind of stuff, you look at them carefully, they've got ethanol in there because it helps dissolve drugs. So is somebody, you know, is having some

Mad Dog 20/20 with their TB drugs, or shortly thereafter, you know, they might have actually improved absorption. But that's probably going to vary drug to drug, and it might have no effect. So some of the illicit drugs are obviously in the narcotic or opioid class, and those drugs will be affected by Rifampin. One way to really bring somebody around is not just Narcan, but give them some Rifampin and that will blow through their methadone or heroin and they will perk right up after that.

I just want to make a quick point is that, you know, our lab here is a nonprofit lab, and my salary is in no way tied to the clinical laboratory here. I convinced the College of Pharmacy to have a clinical lab. Obviously most colleges of pharmacy don't have a clinical lab inside them. But I just wanted to make that point that, you know, whether we do one sample or a billion samples, my salary is completely unaffected.

That means that the kids still go to college, is that correct?

Happily, they're done.

What is it, either college or is -- you know, because you didn't go to college, right, Chuck?

No, I just made this stuff up.

Yeah, well, thank you. Hey, we've got to stop here, but I want to first thank you, Chuck. I mean it's just simply, simply fantastic. I can't thank you enough. One thing though I did really notice, Chuck, is as you -- - you know, initially, when we put up the question about Super Bowl, Denver was way, way, way out in front. The more you spoke though, they're falling, you know. So I think you're having a negative influence on the Denver's prospects for winning. I mean, I hope that's not true.

Well, truth be told, I was rooting for the Patriots, because I'm from Massachusetts originally.

You know, being a -- I won't even go there. But, hey, look, the bottom line comes down to, as you guys know, I mean, pharmacokinetics play such a large role, and the whole issue of drugs and absorption play such a large role in our treatment of TB patient. And I can't say this enough, over the years I've really relied on Chuck and his colleagues to help us out in so many situations. And, Chuck, I really mean this, there's so many patients who really owe so much to you, and there's so many of us who owe so much to you. And I really want to thank you so much.

And I want to emphasize to everybody out there that Chuck is available. I mean, so if you guys have a difficult case and you think pharmacokinetics is becoming an issue and that you may need to do drug levels, I really highly recommend that you call Chuck. And you can get Chuck on the information that's up there or through a 1-800 number. That's 1-800-4TB-INFO. Yes, that's 1-800-4TB-INFO. Other than that, you know, Chuck, I can't thank you enough. And we all want to thank you for everything you do. So, hey, guy, great work. We really appreciate it.

And this is Lola from SNTC, and I want to say thank you to both Dr. Peloquin and Dr. Ashkin for being on the webinar. So thank you both very, very much for your participation. I know this is hard work, and we had some glitches here and there, but thank you both for making it work.

Also, I want to remind everybody that the evaluations will be sent out by close of business tomorrow. So please be on the lookout with your e-mail. If you have not submitted your e-mail, please do so in the e-mail pod below the presentation screen. Also, there are handouts available in the pod handout, as well as on our website under "What's happening." And I want to please wish everybody a nice, warm day. I know it's cold all over the country, so stay warm. And thank you, again, to everybody.