U.S. FOOD AND DRUG ADMINISTRATION

National Center for Toxicological Research

Science Advisory Board Meeting

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P R O C E E D I N G S (8:00 a.m.)

Agenda Item: Welcome and Overview

DR. MENDRICK: Miki Aschner, who's normally our chair was actually stuck in New York City yesterday. He's flying in today. Greg Lanza has been nice enough to step in and chair part of the SAB Forum and I thank him very much for that.

(Housekeeping comments)

DR. LANZA: Thank you very much, Donna. Thank you to everybody who's come to this meeting of the NCTR Scientific Advisory Board. I think it's best if we just go around and introduce ourselves very briefly.

(Introductions around table)

I've been asked to keep us to a schedule, so please work with me. Don't be offended if I say, okay, cut it. With that, I'll turn it over to Donna to talk to us about the conflict of interest.

Agenda Item: Conflict of Interest Statement and "Housekeeping Items"

DR. MENDRICK: Thank you, Greg. This is Donna Mendrick. When you when you speak, also introduce yourself. We need that for the transcript. So good morning. I'm Donna Mendrick, the Designated Federal

Official. And we'd like to welcome everyone to the NCTR, Science Advisory Board meeting. We appreciate the time and diligent work of our board members in preparing for this meeting and for their forthcoming deliberations. I and the Board wish to thank the FDA Regulatory Centers and NIHS for their participation in this meeting and my NCTR colleagues for all their efforts preparing for this meeting.

As a DFO for this meeting, I serve as a liaison between the Board and the Agency. I am responsible for ensuring all provisions of the Federal Advisory Committee Act, FACA, are met regarding the operations of the Science Advisory Board. Also, in my role, a critical responsibility is to work with appropriate agency officials to ensure that all appropriate ethics regulations are satisfied. In that capacity, Board members are briefed on the provisions of the Federal Conflict of Interest laws. In addition, each SAB participant has filed a standard government financial disclosure report.

We have a full agenda, yet strive to ensure adequate time for the presentations, public comments, and Board's thorough deliberations. This special note for all presenters, board members, and other participants, please

speak into your microphone and identify yourself and be sure to turn your microphone off when you're done.

Pursuant to the FACA, we will have a public comment period today offering the public the opportunity to provide comments about the topics being discussed before the Board. We have one person already who wants to give a public comment. I would like to add that during presentation discussion, if board members require greater clarification on an issue requiring participation of attendees in the audience, they may request some information during the meeting through the chair or myself.

In accordance with FACA minutes of this meeting will be prepared, as will a transcript. Both will be posted to our external website. So please remember, this is a public meeting. And in closing, I wish to thank the Board for their participation in today's meeting. Thank you.

DR. LANZA: Thank you. Dr. Slikker.

Agenda Item: State of the Center

DR. SLIKKER: First of all, I want to welcome all of you to a Little Rock, and I am so appreciative of you being here to supply information and new ideas and new insights into the NCTR and the FDA and how to utilize our

facilities and quality employees to the maximum for FDA purposes.

It really is great to have our very experienced and esteemed group here of outside experts to give us input in the Science Advisory Board manner. We really appreciate all of you being here today and our representatives from the various Centers in RoA of FDA. It's always a pleasure to have you here to learn new information about ideas that you have not only progress on ongoing relationships, but also new ideas about things that we can tackle together in the future. So, we really appreciate that input. And of course, the audience and the presenters from NCTR for preparing for this particular meeting. It's really a pleasure to have you all here to discuss the progress that we've made during the last year, look for new opportunities for interactions, and to build on the science base for FDA decision making.

So, one thing that I want to do is just really talk a little bit about the unique character of NCTR. It was really designed back in 1971 to be a place where we could bring together resources and collaborate on issues. And this included input from industry as well as input from other government agencies, especially the other Centers of FDA and also with academics to build

collaborations and to tackle problems in a group way. And I think that insight has continued to be a driving force for the NCTR and its support of FDA decision making.

One of the things about it is that we do have the vision, of course, of bringing together activities and collaborations that would involve all these entities that I spoke of. Also, the idea of training and developing innovative science solutions to problems that face the FDA. And the mission really is to then generate data for FDA decision making and to develop new innovative tools and approaches, put them in a format that will be useful to FDA, and to use those to generate data for FDA decision making.

We do this with a structure that's noted here in terms of the Office of the Director having several different boxes underneath it. The first row is one in which we have information from the various kinds of management groups, and this includes individuals interested in budget, also formulation of new projects for the next year. It includes also regulatory compliance and risk management as well as those having to do with regulatory activities. And so, these are the management side and it's a relatively low percentage of the total population.

The real business end of the NCTR are the Divisions located in the bottom boxes, which includes Microbiology, the Division of Genetic Molecular Tox, the Division of Biochemical Toxicology, the Division of Systems Biology, that of Neurotoxicology, and also Bioinformatics and Biostatistics. And you'll be hearing from each one of these particular division directors throughout the next day and a half. So that's our organization and we appreciate that leadership and also the leadership from Rear Admiral Denise Hinton, who is the Chief Scientist within the FDA.

Now, let me just talk a little bit about the staff at NCTR. You can see we have 290 or so government FTEs involved. This includes research scientists, support scientists, as well as administration. And we also have onsite contractors. And this is largely due to the need that we have for animal husbandry. The facility is taking care of the physical plant as well as the pathology group. And all those constitute the contractors that are on site. And then, of course, we have ORISE trainees at a number over 50. And this is really important to us because training new regulatory scientists is one of the goals that we have within the NCTR and FDA.

So, some of the research goals really can be spoken about in this way. Certainly, advancing knowledge and the tools required to support regulatory decision making are very critical for the NCTR. And this, we feel, is important not only to drive NCTR forward, but to help the Agency make those science-informed decisions. Also, to enhance collaborations across the various Centers of FDA. And that's why it's so enjoyable to hear from each one of the other Centers' and ROA about their activities during the last year and new ideas for the future. And then also to promote global interactions in a way in which we bring the FDA community together with those regulatory agencies and individuals working in the regulatory environment all around the world. And we'll talk a little bit more about that later.

So, let's just get down to sort of three simple top accomplishments for 2018, 2019. Part of these really involve this idea of partnerships within the FDA of building partnerships between the Centers and also with the other institutions that are government agencies within the US. And also, the idea of advancing regulatory sciences in general. This is developing newer technologies, preparing them for being useful to FDA regulatory decision making, comparing them against

guideline studies that are so critical to making those firm scientific based decisions. And then also, of course, as I mentioned, advancing regulatory science in a global fashion.

Now, just to get down to some examples, some of the partnerships that have been developed with CDER, Center for Drugs, you realize that we have many interactions with them and some of them can be outlined here, including the work that we're doing with opioids. Obviously, this is a crisis across the US and there are many different studies going on to try to understand this and figure out more precisely what the impact of opioids could be, especially during development and prenatal exposure to opioids. And so those studies are ongoing.

Also, in the area of pediatric anesthetics, this is really looking at long term exposure to anesthesia for some critical surgeries that are necessary, but are there impacts of that, is one of the questions that we've been examining with CDER over the last decade or so, and that continues through this day. Also, MOU continues looking at interests including sunscreen ingredients and other non-prescription drugs. This has been a great relationship with CDER and will continue into the future as well. And then also developing new methods to look at various kinds

of bacterial contamination, especially in pharmaceutical products. So those are some examples of work that is ongoing with CDER.

With CVM, the antimicrobial resistance and the human microbiome is really a critical issue. And those studies have been ongoing for some time as well and will continue. And also, the idea of looking at veterinary drug residues in food on the intestinal microbiome. That is, how do they affect the microbiome, which then has consequences downstream. So those are just some examples with CVM.

With CFSAN, we have many different things going on. But just one example is the detection of microbial contaminants and this is really important in terms of tattoo inks. There have been some identified with unfortunately they contain some bacterial contamination, and this has been worked out between CFSAN and NCTR as far as how to assess those kinds of contamination.

Also, with our colleagues at Office of Women's Health, we've worked on precision medicine ideas, especially in the studies of triple negative cancers in African American women. This kind of study really focuses on male/female differences and oftentimes on ethnic diversity issues that have to be addressed.

And then in a general way, we've had many public workshops, some of them looking at various forms of artificial intelligence and bioinformatics tools. And this is especially relevant to sequencing quality control. This is going on across the entire agency. And many of these activities are coordinated by NCTR.

So, looking at the work with CTP, which is one of our partners, we are very proud of this relationship with CTP. It's been an excellent opportunity to build capacity within the FDA and to do it in conjunction with our colleagues at CTP, the Center for Tobacco Products. And part of that has to do with inhalation facilities that are available. And these studies can be done to look at nicotine pharmacokinetics or NNK and other kinds of possible inhalants that are associated with tobacco products.

Also looking at alternative models and the idea is using an in vitro 3D air-liquid interface. This allows you to do this with the human airway cells and to evaluate the possibility for toxicity and inflammation in this in vitro setting. And then also modeling and predictive toxicology. And this is the idea of using physiologically based pharmacokinetics models to evaluate agents that are associated with tobacco products. And these studies have

been ongoing for some time now and will continue in the future. But we really do appreciate the close collaboration with the scientists within CTP in building this capacity within the FDA.

Let me also talk about our partnerships with some of our other agency groups. And this one is with NIEHS and the National Toxicology Program, NTP. This interagency agreement has been actually going on for over 25 years now. It's really built great capacity within the FDA and also allowed the National Toxicology Program to talk about its impact on public health and clinical care, in particular. Some of the projects that are ongoing in conjunction, of course, in each case with other Centers, one with CFSAN is looking at the metabolism and toxicokinetic studies as related to arsenic. And these toxicokinetic studies are really groundbreaking in that they're describing exposure modalities to arsenic that hadn't been covered before.

In terms of a relationship with CDER and CBER, NTP is supporting studies that we're doing on pegylated compounds. This is really an exciting one because it brings together two other Centers as well as the NCTR and NTP working together. Another one really focuses on the evaluation of brominated vegetable oil and this is done

with CFSAN, obviously an issue having to do with a food source of contamination. And then we also have various work going on with CFSAN with lumbrokinase and other agents that are known to affect blood in several different ways.

Also, with CDRH and the National Toxicology Program, we're looking at disease related toxicity effects of inhaled compounds and doing this in an in vitro mode. And this this kind of approach is useful not only to NCTR, but also to NTP as well as the Center for Devices and Radiological Health. And then finally, one that we're doing in conjunction with NTP, really looking at the role of microbiome and how you incorporate that into toxicological studies. Important to look forward in this mode and understand how we can best use the microbiome data to supplement and interpret traditional toxicology type studies. Those are just some examples of those partnerships that are going on between another agency, NIEHS, and the FDA.

So, let me just briefly then talk about some of the methodologies that we've been working with and developing over the last several years. Certainly, we've already talked about some of the role in safety assessment that NCTR helps to lead and move forward for the Agency.

Biomarker development and also validation is really a key feature, including everything from imaging, which is the next topic on here, using imaging as a biomarker in a minimally invasive way, to looking at all sorts of genomic biomarkers as well as others.

We also have been doing a lot of work with 3D models and stem cells. This is really critical not only to evaluate the use of human stem cells to make toxicological comparisons, but also to evaluate stem cells in general as a model to compare against in vivo technology and making sure that they're reflecting something that can be useful for decision making. We've already talked about the microbiome. And a little bit on precision or personalized medicine, by developing and understanding the use of new biomarkers, one can move the field of precision medicine forward. And so that's one of the approaches that we've been using there.

Nanotoxicology is an area that's really been developed within our Nano-Core Center, which is a joint interaction between the National Toxicology Program and also ROA within the FDA and NCTR are working together. But we also, of course, share this facility and capability throughout FDA with the other Centers. It's really been a force because it has some of the best equipment and

technology as well as personnel to address these issues in the nano area and to also be a framework for having meetings that bring people together to talk about nanotechnology and how we can evolve safety assessments for nanomaterials in the future.

Inhalation toxicology is done in conjunction with the Center for Tobacco Products is really a key issue here. This area has been developed not only for whole animals, but also for cells and culture. So, it's a real step forward in understanding the best ways to do inhalation toxicology across the FDA.

We've talked a little bit about pharmacokinetic and pharmacodynamic modeling. This biological modeling is really key, not only to extrapolate then between species, but also to understand more fully how you would extrapolate from in vitro settings to in vivo settings. Bioinformatics tools and artificial intelligence is another area that we've really been advancing. This has to do with the idea of maximizing the data that we have available to us, looking at data in a whole new way to give us information about safety assessment and to develop new tools that can be useful to the FDA decision making process.

And then, of course, we have already mentioned the idea of our ORISE program and other training opportunities. We also have a summer undergraduate program for training as well as the postdoctoral fellowship program. All these work in a way to generate scientists that understand and are well versed in regulatory science for the future. So those are just some of the areas that we've been working on over the last few years and into the future.

Let me just give you a couple examples. This one has to do with advancing FDA regulatory science and really the idea of how you develop tools that can be useful, especially in the bioinformatic area. So this Review-to-Research and Return is an idea of working with reviewers, understanding some of their needs that they have to make decisions and the timelines that they work under, and developing tools that can be helpful for them to accomplish your goals in a more efficient way.

So, these interactions have been very useful. Some of them include Data Analysis Host Systems, as well as being able to track progress on INDs and NDAs, et cetera. The other way is to really upgrade systems that already exist or make them more user friendly and more efficient for the reviewer. And this is going on a routine

basis between our Division of Bioinformatics and Biostatistics and the other Centers of FDA, including CDER.

Other areas, as I mentioned, precision medicine certainly is a collaborative process and bioinformatics plays a role here to really understand what sort of biomarkers are most useful to move precision medicine forward. And then as I mentioned, artificial intelligence and deep learning methodologies are just another example of how we've been using new tools and developing new processes that can help make decisions in a more rapid way for FDA.

So, let me then turn to a couple of the last items I wanted to emphasize, and one of these is the progress that we're making on maternal and children's health. We developed the Perinatal Health Center of Excellence about two years ago. This is an FDA-wide organization that received funding last year in 2019. And this funding then goes to have the opportunity to perform experiments based on protocols and proposals from all the different Centers or ROA, that come in and are evaluated by a committee of experts represented from each Center and ROA of the FDA. And decisions are made about which will be funded for the future.

This has been moving forward very nicely. We have the opportunity to look at this because we think it is important to not only maternal/fetal pairs, understanding that relationship between the maternal and the fetal system, but also to evaluate preterm effects and infants where we can study them as possible using animal models, cells in culture as well as clinical studies, and also provide this ability to address these kinds of needs across each one of these Centers, ROA included.

Now, what some of the steps that we've made is first to get funding in the FY-19 budget, as I mentioned. Then we were able to receive proposals with a call for proposals and we had opportunity to review 22 proposals in this area of perinatal health. And this includes everything from the maternal health, premature, new infants, as well as childhood. It's a broad definition to indicate that we're interested in that total framework and to be able to provide support for experiments that focus on this entire area of perinatal health.

So, we were able to evaluate 22 proposals and we found 14 of those were of high quality and they were funded last year. Those will continue on into this year. Each one is a two-year project. Also, we were able to hold a workshop where we brought all those individual

researchers together and their colleagues and presented their work at about a year into the projects, at a nice White Oak meeting opportunity. And this really provided them with a good framework to describe their work and the progress that they made in their work during that first year.

And then of course, this year, we went out again with a call for proposals, received 10 new proposals. And of those, we were able to fund three with the funds that we had available at this point in time. So right now, then we have 17 projects moving forward in this area. And next year, as those first-round projects are finished up, we'll be able to fund a whole new group of projects at that time. So I think this is moving forward very nicely and also is bringing together researchers from each one of the other Centers in ROA to work on these in a group way and to really understand more fully how we can have positive impact in this area of perinatal health.

Let me just turn then briefly to one of our last topics, and that is looking at the global outreach and we're doing this in a number of ways. But one of the ways that we've been building over the last 10 years or so is using this Global Coalition for Regulatory Science Research to foster development and to interactions across

various countries. We have now, working together, 10 different countries and the EU. And you can see the countries listed here, we meet on an annual basis and have an international meeting at that point in time, a global meeting. But we also meet online and with conference calls during the year.

But the idea is to not only support an annual meeting called a Global Summit of Regulatory Science, but also to foster collaborations by not only understanding what collaborative events could be most useful to the agencies around the world, but also what areas of training can be available through online services that can be useful to each one of these countries as well. So, we feel this is a real opportunity to focus on research. We don't focus on policy or inspections. We focus on the research side that supports regulatory science in a global fashion.

Just a couple of examples. In 2018, the Global Summit on Regulatory Science was held in China and it was on the risk benefits of dietary supplements and herbal medicines in the era of data science. And this writeup on this particular one is in progress now. We hope to have that published in the next couple of months or so. The most recent one was held just in September in Italy. It was supported not only by, of course, the Global

Coalition, but also by our colleagues there at the JRC. The idea there being that it really focused on nanotechnologies and nano plastics, which is really an exciting area and one in which we revisited from 2016 but found this one to be very energized with participation from 34 different countries with over 200 individual participants. So, a very successful meeting held there, Northern Italy that supports the whole concept of learning more about nanomaterials and nano plastics.

The next one will be held as sort of the 10th anniversary meeting with a theme of Emerging Technologies and Their Application to Regulatory Science. It'll be the end of September, 28th through 30th, in 2020, right there in Bethesda, Maryland, at NIH in conjunction with NCATS. So, we're very excited about this meeting that's coming up. And certainly, invite all of you to put that on your calendar and hope that you will attend and also present. We offer the opportunity not only for standard presentations but also posters.

So, this just gives you an idea of some of the activities that are moving forward in this area of regulatory science in a global way. Let me just finish up by just saying that we feel we've made some progress during the last year and look forward to making more this

coming year. Certainly, some of the activities have been on trying to understand more about how we can coordinate with the other Centers and prioritize action plans to move things forward.

There are some areas that we think are especially ripe for consideration: cell systems, both stem cells and others using human cells and animal cells for a comparison. It's an area that we think is really one that will move areas forward. Emerging technology, we've talked a lot about that, everything from bioinformatics tools to new tools using stem cells and imaging approaches. Of course, modeling is a big part of that. There will be laboratory animal studies. One of the few places in the US Government where guideline studies can be performed is at the NCTR. And therefore, those studies will be the framework oftentimes for a comparison with new technologies and also generate data for FDA decision making.

The bioanalytical skills in chemistry continue to increase. And those are used for basis for modeling and for comparison across species. And also, we talked quite a bit about informational sciences, artificial intelligence, and how those new bioinformatics tools are being so useful not only for developing new technologies, but also for

understanding more about omics technologies and how to utilize those data in a safety assessment mode. So, we feel like the collaboration across Centers is key and that we want to continue to enhance that over the next year.

So, let me just finish then with some questions for discussion. And you know, really what we want to do here is just get you thinking more broadly perhaps about what sort of things that we'd like input on. And you can certainly use these as some of the things that you might want to respond to in your written report, as far as the Science Advisory Board members, but really what sort of animal models can be better utilized for preclinical decision making and what tools might help to enhance that. Also, the idea about regulatory approaches. Are there some that can be replaced with emerging technologies? When should that occur? What are the validation steps to make sure those new technologies reflect the best science?

And then further to really think about the use of artificial intelligence and in silico approaches, how can we improve those to make them more useful to FDA decision making? And then also the idea of pharmacokinetics in the in vitro to in vivo extrapolation, how can we do that in a more concise way? So those are just some of the things that we've been thinking about.

There may be many others at the Science Board will also come up with, of course, as they see the various presentations. But we want to get you thinking early about some of the things that we'd like to get some feedback on.

So, with that, I will close and just thank you once again for the opportunity to present. And for all of you being here, we really appreciate your great collaboration and the questions and kinds of inquiries which you'll make, as well as the ideas that you'll bring forward, so that we can use those to optimize the safety assessment of various products within the FDA. So, thank you all very much.

DR. SLIKKER: I'd be happy to take any questions if you have some now, or we can carry on with the agenda as set. All right, yes, please, Ken.

DR. RAMOS: Thank you for that update, Bill. Your portfolio is quite impressive, so congratulations for that. I have a couple of questions. How normally does the NCTR transfer knowledge outside of the Agency? How is that sort of orchestrated?

DR. SLIKKER: Yes. I'd say at least in a couple of ways, Ken. That's a good question. One is sort of the traditional way in which the NCTR and FDA likes to have their data peer reviewed and published in the peer review

journals. And so that's one of our goals is to get it that way. The other way, of course, is to have these meetings, either they're standard meetings that are held by societies where information is transferred in that fashion or some of these specialized meetings like the Global Summit, where we bring together individuals from 10 different agencies around the world in the EU and discuss the issues there. And then out of those come a report that is an annual report of that meeting contents. So, we use these various mechanisms, both the traditional peer review publications as well as specialized meetings to get information out. We think that's really critical and one in which we rely on sort of the traditional sources.

Now, there's also lots of group activities within the FDA to transfer information back and forth from the various review centers and ORA to the NCTR and back and forth. And so those meetings are set up for us to transfer information as well. But the outreach to the rest of the world is through your publications and through your meetings where we can release information in a timely way.

DR. RAMOS: Do you guys put together an annual or biannual report that captures all of this information? And if so, can you share a copy with us of that?

DR. SLIKKER: No, I appreciate that. And of course, we do have an annual report that is available online. It is available through our website. It summarizes the 170 some active protocols in terms of title, PI, and objectives. And so, we use that kind of information to make sure that everyone is aware of the breadth of our research. Yes. And from that, of course, you can ask more questions if you need to, but it certainly gives you an overview of the different studies that are ongoing within the NCTR/FDA.

DR. MENDRICK: This is Donna Mendrick. Our most recent annual report is actually in the book under Item 10.

(Off mic comment)

DR. SLIKKER: Yes, please, Greg.

Dr. LANZA: Dr. Slikker, I think now is as good as any. I noticed several times you mentioned AIAI and I wanted to make a comment, allow you to respond. I've become much more involved in AI and at a very high level with very strong people. And so, one of the things that I noticed is that you're interested in AI primarily - if I could generalize - for workflow, operational, going through adverse report, so forth. But I think that NCTR has a mission in AI that's almost unique because you cross

all the different Centers, but that is that AI has the ability to help in the toxicology in two ways. One is to take information, particularly that you already have at the FDA in specific areas and predict how that related - train, if you will - to the adverse effects that we're seeing. And in doing so, you create a neural network which has nodes that would help you to identify what were the key issues.

But even greater is the adaptive, particularly building on that as you go forward, because so many topics, so many specialized interests. And I think that AI here could help in recognizing and predicting a toxic project, toxic things that wouldn't be seen unless your patient numbers were in the millions or even in the hundreds of thousands, which really doesn't come through in helping these different agencies predict that. And it would be not only using all the information you have, diverse information to help understand what things might be toxic so you can focus on those issues, see if it's true or not, but also, how it got to that. And that relationship between how the machine understands questions helps to overcome our bias.

And with the position of NCTR are at the core, this may well be one of the most important new things I

think that you need to gain. And you need to bring in not just people at a basic, just started to work in the area, but the pros pros. But you have all the data. You have all the applications. No sponsor has all the information that you can bring to bear to pick out issues.

DR. SLIKKER: Well, I appreciate those comments, Greg. And certainly, we have invested heavily, including a Division of Bioinformatics and Biostatistics that focuses on AI, as well as various tools that have been developed in conjunction with the other Centers, including FDALabel and a few other ones that utilize data that's publicly available. I know that everyone would like to get to some data that's not publicly available, but that is protected very carefully by FDA. But certainly, publicly available data can be used in such a way as you suggest and certainly has been as exemplified by FDALabel and some of the other tools that have been developed between the Regulatory Centers and NCTR.

DR. LANZA: If I may follow on, I wouldn't even be interested in the publicly available data. It's the data that was in the sponsors' applications that you have access to help you isolate what key issues might be coming forward to train and then to use that to help you evaluate and even predict toxicology issues that you can then focus

on better. But it has to start at the beginning, and this is the best place they have the data. But you're in the catalyst position of all the different Centers. And that's what I think really needs to happen. Just my opinion.

DR. SLIKKER: As I mentioned, there are definite limitations to that and there are also some opportunities. So, they have to be evaluated as such within the rules and regulations of FDA. Yes, please. Yes.

DR. GANEY: Again, thank you for that update. I have a really quick question about the Perinatal Health Center of Excellence. I notice that there were far fewer -

DR. MENDRICK: Please introduce yourself for the transcript.

DR. GANEY: Oh, I'm sorry. This is Patti Ganey from Michigan State. I noticed that there were far fewer applications the second year. Is this a difference in the RFP or do you think you're saturating the field, or do you expect that trend to continue? I'm just curious about that.

DR. SLIKKER: Yes, well, thank you for noticing that. It really was based on the RFP indicated very clearly that we had limited funds for this second year. The reason is, is that we have to utilize funds on a yearly basis within the federal government. And we utilize

all the funds in the first year to fund those 14 protocols. They're two-year projects, so they run the second year. So, we only have the same amount of funds the second year and therefore, we only had a small residual left that could be used for new projects in the second year. We warned everybody about that in advance. Therefore, fewer people took advantage of the opportunity and we were able to only fund 13 rather than 14.

Now next year, we'll be back in full funding mode again. So, we're trying to get additional revenues to sort of level out that issue. But that's how we're in that situation. It has to do with the utilization of funds completely in one year to fund those projects and start them up. And most of them are two-year projects. Thank you. Yes.

DR. COSENZA: Mary Ellen Cosenza. I have a question on educational opportunities. So, you have a unique opportunity in that you have laboratory animal studies that you can run here or at NCTR at the site. And I know you can actually graduate with a Ph.D. in toxicology in this country right now and never actually handle an animal or run a toxicology study. So, we know we have people in the industry, but also obviously in the FDA who are reviewing these studies for sponsors but have

never actually run such a study. So, are there educational opportunities for toxicologists from all the other divisions to come here and actually do like an internal training and have that opportunity to do that?

DR. SLIKKER: Well, most certainly that is available. Many of the other Centers do have animal facilities and run studies with animals of their own. But at NCTR we do sort of emphasize that as an option and an opportunity and provide that kind of training. And certainly, we have good exchange not only between the principal scientists and the various Centers, but also with many of these students who are in training. And oftentimes some of the students after they finish their three years or so of studies here as a postdoctoral fellow may find employment in the other Centers, so it works out to the idea of training individuals can be available within the FDA or can be available to other government agencies or be available to industry and academic facilities as well. So, we've trained individuals that have gone into all those different areas and we think that's an important role for NCTR. Yes, please.

DR. STICE: Steve Stice. Bill, congratulations, and to all the scientists on the progress you've made. Getting back to the Center of Excellence a little bit, now

that you're in your second year coming on your second year, you must have some metrics for success and what you determine as success. Can you expand on those?

DR. SLIKKER: Well, at this point in time, we're using that annual workshop to evaluate progress. And what we were very proud of is that even though it was a difficult year last year for the FDA and many federal agencies, that they were still able to start their projects, get all of their approvals that they needed, and actually generate data in the first year. And so, we got a chance to evaluate some of that data. And the real progress is going to be based on their annual report that'll be coming up at the end of the second year. And hopefully from that, manuscripts, and publications. So, we're using those traditional markers as well as the annual workshop to assess progress. And they do have the opportunity and responsibility to provide reports not only annually, but also semiannually as to their progress. John-Michael.

DR. SAUER: John-Michael Sauer, Critical Path Institute. Bill, I really appreciate you going through all the different collaborations that you have. I mean, a lot of them are across the Center. There's one external one that you discussed with NTP, but are you pulling other

stakeholders in? Because I look at your list of areas you want to advance scientifically. And of course, there's other groups out there, other stakeholders that are working on this as well. Do you have plans to interact with those stakeholders and how does that happen from an NCTR standpoint?

DR. SLIKKER: That's a really good question. And we use several mechanisms. One of them is the opportunity to actually provide contracts to outside sources. And it depends on, of course, availability of funds. But the idea is that you can do a broad area announcement and do a contract with an outside source. There's also the use of proper research development agreements, of which we have several of those running. And those can happen between the FDA and university or the FDA and industry. But everything is defined very carefully, and of course, goes through both ethical as well as scientific review to establish those. So, I did not get into those details, but we have those mechanisms and we use them routinely.

All right then. Well thank you all very much.

DR. LANZA: Thank you very much. Let me introduce Dr. Susan Felter. She's the Chair of the Subcommittee that reviewed the Division of Genetic and Molecular Toxicology.

Thank you, Susan.

Agenda Item: Subcommittee Review of Division of Genetic and Molecular Toxicology

DR. FELTER: So, the full report of the Subcommittee review is in Tab 6 in the notebook. And I'm presenting on behalf of myself and Dr. Michael Aschner, who's not here today. And then we had three subject matter experts who joined us for the technical review: Dr. David Eastmond from University of California Riverside, Dr. Mark Fielden from Amgen in California, and Dr. Ofelia Olivero from the National Cancer Institute. And most of the written reviews on the project specific reviews were done by these subject matter experts. The review was conducted over two half days, which included about an hour and 15 minutes for viewing quite a few posters, and that allowed the NCTR, the DGMT to expand the amount of information that they were able to share. We didn't all get to see all of the posters, but we went to the ones that we had primary expertise in. So overall, we really enjoyed that as a new format that was used, but it may have been a little bit too much (gap in audio file) period of time.

The project overviews were divided into three thematic areas, and so reviews for each of those three areas were written by the assigned experts. So, the first reviewer was David Eastmond and he reviewed Theme Number

1, which was current research supporting regulatory acceptance of the Pig-A gene mutation, that research coming out of Dr. Dobrovolsky's lab. The second was new approaches to genetic analysis, and that was Dr. Parsons, Revollo, and Chen. The primary reviewer was Mark Fielden. And the third was new biological platforms for evaluating genetic toxicity by doctors Wang and Petibone and the reviewer there was Ofelia Olivero.

So, I'll start by saying that for each of the three focus areas, the Subcommittee found that the DGMT research is highly relevant, highly impactful, and consistent with its mission and that of the FDA. So overall, it was a very positive experience and I think the reviewers unanimously had a lot of very positive things to say in the course of the review. So, the structure of our report is to go through each of the three theme areas and then some general comments at the end.

So, the review of Theme 1, again, that was current research supporting regulatory acceptance of the Pig-A gene mutation assay. So, this assay detects mutations in the X linked Pig-A gene. And it was developed using hematopoietic cells of several mammalian species, including humans, and has been shown to be useful in non-clinical safety evaluations for detecting potential

mutagens and carcinogens. And as an in vivo gene mutation assay, it's seen as a valuable test which can be used to follow up positive in vitro results and thus should be able to fill a critical void in current regulatory testing schemes.

So, one of the big advances here started - I should say, this report, the review was held in March of this year, was postponed from December and the report was finalized in May. So, this information, you might want to add a year to these timelines. But about five years ago, a proposal to create a regulatory compliant test guideline for the Pig-A assay was submitted to OECD and this was done by a consortium that was led by DGMT scientists. And while it was generally viewed as promising, there were some concerns that were raised at the time by the OECD reviewers. And so, Dobrovolsky, Heflich and other members of the DGMT, in collaboration with outside stakeholders, took the lead in addressing the concerns and then submitted a detailed review paper on the Pig-A gene assay to the OECD.

And at the time of our review, work was continuing on a retrospective performance evaluation of the assay that was going to be submitted to OECD in time for it to begin its formal review. And it was anticipated

that the test guideline would be approved by 2021. So overall, the Subcommittee felt that the work led by the DGMT conducted on the assay in preparation for this OECD guideline, which is a really important step forward in its use by regulatory agencies, that work done by DGMT, provided critical information to support the validity of the assay and really represents a valuable contribution to the genetic toxicology and regulatory fields.

There was also work that was highlighted for DGMT scientists who were beginning a study collaborating with University of Arkansas researchers to determine erythrocyte Pig-A mutant frequencies in cancer patients before and after undergoing cisplatin-based chemotherapy. And in conjunction then with in vitro and animal Pig-A mutagenesis studies on cisplatin, it was felt that that study, which I think is ongoing now, has the potential to confirm the usefulness of rodent assays for detecting mutations that are relevant for human risk and also to provide insights into the potential risks for humans receiving this type of chemotherapy. So overall, this again highlights the relevance, timeliness, and impactfulness of that research.

The second theme was new approaches to genetic analysis, and there were three projects discussed under

this theme. And again, Mark Fielden was the subject matter expert for the Subcommittee. That first project was led by Dr. Parsons and this was cancer driver mutation-based biomarkers for cancer risk. So, this research will help to improve our ability to predict future cancer risk and potentially help improve the translatability of animal data. The DGMT are encouraged to further develop the expertise in error-corrected, next-gen sequencing approaches, and apply their experience in validating new mutation assays such as the Pig-A assay to this emerging area of mutagenesis research. And just a side comment that when you're successful, this is what happens. You're asked to do more. So, Dr. Parson's work has helped to establish the importance of variability in cancer driver mutations as a marker of risk.

And to expand the significance of this research, it will be important to address what set of driver mutations will be important to assess for specific tumors of interest and how these may behave in non-clinical models. The DGMT should continue to avail themselves to both non-clinical rodent models and clinical samples to further test these hypotheses and improve our understanding of the translatability of rodent data. Adopting state of the art approaches for ultra-rare

mutations will be key to advance this field. So, the DGMT should consider developing experience with other error-corrected sequencing approaches as they become commercially available as well.

The second project was by Dr. Revollo and this was assessment of clonal whole genome sequencing for detection of gene editing induced off-target effects. So, this research is focused on establishing the unintended genetic side effects of CRISPR based therapeutics and thus is important for the safety, assessment, and regulation of these products. And it addresses a significant gap that requires new tools since existing gene tox assays do not fully inform the potential risks of CRISPR based therapeutics.

It is acknowledged that detecting rare somatic variants such as insertions or deletions can be difficult to detect with short-read sequencing technology and may require single-cell cloning and expansion. Careful attention to artifacts that may arise during the expansion phase will be required. One limitation of this approach is the need to source individual cells, which prohibits application to certain cell types. So, it was recommended that the DGMT consider exploring methods that can be readily applied to a variety of tissues, models or species

and can be incorporated into existing tox studies where single-cell cloning is not readily feasible.

And it was also recommended that the use of mammalian models should be prioritized as feasible since the evaluation of base editing and E. coli and germline mutations in C. elegans may be problematic from the perspective of human relevance. I don't know if problematic is necessarily the right word so much as having questions right now that are not answered regarding human relevance.

The third project presented by Dr. Chen was assessment of mutagenicity of nanomaterials using whole genome sequencing. For this project, the efforts to study the mutagenicity of nanomaterials is challenging owing to the physical attributes of these particles and limitations of the Ames assay. So, the proposed approach of evaluating mutagenesis in vitro using whole genome sequencing of clonally selected cells may provide an attractive alternative. The recommendation was that the DGMT should consider first establishing an in vitro model with appropriate negative and positive controls to characterize the system and establish the methodology prior to investigating and interpreting effects with new compounds

such as silver nanoparticles. So, some focus on the fundamentals of the assays.

Initial characterization of the model will also facilitate an understanding of the strengths and limitations of in vitro models to predict how the results may translate and how it compares to "gold standard" approaches using the Ames assay, the mouse lymphoma and/or in vivo endpoints.

So overall, for this session, the DGMT researchers were encouraged to continue to evaluate and develop the next-gen sequencing work that's already ongoing to understand the exact base changes involved with mutation. This will help determine how diseases progress through the induction and expansion of mutations and to leverage mutations signatures to associate cause with effect. So, in particular, exposures with disease causing mutations. This technology promises to significantly advance the means by which in vivo, mutagenesis is evaluated, and it is anticipated to have far reaching implications for hazard ID and risk assessment. And it will also be important to address potential risks with new therapeutic modalities such as gene editing, which has remained a significant gap.

Two other recommendations that were offered. One is considering the many potential applications of next-gen sequencing to mutation detection, the DGMT should consider what an ideal minimal in vitro and/or in vivo test battery might look like, that would provide the best characterization of chemical induced DNA damage from the perspective of understanding mechanism of action and dose response, and also which considers animal use. In addition, there should be some consideration for how a new model would replace what has already been used rather than just being an add-on to the existing regulatory requirements.

And then the one last comment that was offered was in the area of epigenetics, where it is acknowledged that epigenetics can play an important role in carcinogenesis and in inheritable phenotypes. However, simply measuring changes in methylation state, for example, with the Epicoma assay, may not provide the information needed to fully understand the risk of such changes. And I know the NCTR is fully aware of that and has been working on this for some time. There are some basic biological questions related to the cause versus effect of epigenetic alterations in tumors, and the degree to which changes, quantitative or qualitative, induced by

a chemical can meaningfully contribute to an understanding of risk with any certainty, particularly if we're talking about an in vitro model. So the DGMT should carefully consider the degree to which these basic questions and the role of epigenetics are explored relative to the effort involved and the competing priorities of the Division, as well as the relative impact of any new assays on risk assessment at this time. So again, I think the recommendation there was for some consideration of the fundamentals of epigenetics as it would apply to applied science.

The last theme was new biological platforms for evaluating genetic toxicity presented by doctors Wang and Petibone with the lead reviewer was Dr. Ofelia Olivero. So, doctors Wang and Petibone presented research proposals for new biological platforms for evaluating genome toxicity that were developed in part to address limitations of existing in vitro assays, including tissue cells, specificity of induced toxicities, three-dimensional structure, and metabolic capacities, and the desire that we closely simulate the in vivo environment.

Dr. Wang presented an organotypic human airway tissue model to evaluate genotoxicity. And Dr. Petibone presented a testicular model to evaluate effects on germ

cells. The subject matter experts who reviewed this agreed that the models do help to address the current limitations that were highlighted with our existing assays and that there is potential to improve their capabilities.

So, the bronchial epithelium model presented by Dr. Wang will be used to detect mutations using the Pig-A gene reporter assay with an aim of bridging gaps between in vitro and in vivo outcomes and limiting testing in animals then. So, the method is seen as promising and closer to human scenarios of exposure. There was a word of caution that was offered, which was to emphasize that the 3D cultures need to be biologically relevant and recapitulate micro-environmental factors that closely mimic in vivo situations.

One of the components that should not be forgotten is the extracellular matrix and its 300 interactive molecules. And of critical importance for the endpoints in question are the infiltrating immune cells that in multiple occasions are the producers of oxidative stress and consequent DNA damage.

The model presented by Dr. Petibone seeks to address the complex concept of germline mutations by providing a tool that can help with designing studies, dose selection, time of exposure, and aid in

interpretation of germline mutations before moving on to in vivo models. It is advisable that the model be developed to obtain sperm cells that in turn will provide a venue to address a different set of toxicities associated with exposures and then associate those with endpoints such as malformations, motility issues, and others.

So, the subject matter experts concluded that the DGMT investigators have a challenging but exciting time ahead of them, optimizing the models and moving forward with the concept of diminishing animal testing.

So those were the sum of the technical comments, all of which really highlighted the significant capabilities and advancements of the DGMT scientists to moving genetic toxicology forward, helping to reduce the need for animal testing, and making the testing that we do have more human relevance. I think overall the comments were all very favorable.

There were some overall comments provided at the very end. I'll just hit a couple of the highlights here. One is that it is evident from the history of achievement of the DGMT and the materials that were provided to the Subcommittee that this division provides essential support to the mission of NCTR and FDA overall in the area of

genetic and molecular toxicology. The DGMT scientists have substantial experience with many of the commonly used assays and have been involved in developing the guidelines and are a valuable source of information for the FDA Centers on the performance of the various tests and on the interpretation of the test results.

The DGMT Scientists are highly collaborative and very well integrated into the broader scientific community, including across FDA Centers but also outside of FDA, including professional societies and international consensus-forming groups, where in a number of cases - and I've talked to a couple of these - DGMT scientists have leadership roles in developing that international consensus. The Subcommittee found that the evolution of the Division over time speaks to its agility and focus on meeting the needs of the Agency. The DGMT is highly effective at providing data needed for genetic risk assessment, product safety assessments, while at the same time strategically advancing research to advance the field.

The quality of the science performed by DGMT was found to be outstanding. The DGMT scientists use innovative approaches to solve important issues required to advance regulatory science, and a number of examples

were highlighted during the Subcommittee review. One caution that was offered was just to ensure that where appropriate and feasible, which is not always the case, that the research remains hypothesis driven rather than technique driven.

One area in which the Subcommittee saw the DGMT having a unique opportunity to be influential in moving regulatory toxicology forward is in the qualification process of new biomarkers or models, which is currently quite lengthy. And we heard Bill recognize that as a key area that NCTR is working on as well. So again, the suggestion was that the DGMT should consider how their experience in validating the Pig-A assay could be leveraged to help develop guidelines for how new genetic toxicology tools should be qualified for regulatory use. So, take the gains there and apply them in other situations.

The Subcommittee greatly appreciated what was clearly a significant effort by the DGMT staff to prepare really meaningful materials, including many presentations and posters that were available for our evaluation and discussion. We recognize that this work represents only a fraction of the capability and the achievements of the DGMT. We encouraged NCTR to continue to keep the Division

reviews focused on those projects for which input is specifically sought since it's clearly not possible to do a complete review of a division within one day. We did have some other small suggestions on the logistics that might be considered for how to handle such a large amount of material in one day. And that's pretty much it.

So, on behalf of Miki and myself and the Subcommittee, I did want to say thank you again to the DGMT for really putting together a really meaningful two half days for us. And we found it to be very useful and we hope that the report was as well.

DR. LANZA: Thank you very much. We have just a moment, but before we vote in the Subcommittees to accept the report, does anyone have any short questions to clarify on the SAB? We'll be discussing the response and having discussions subsequently. Hearing none. Why don't we vote by hand? Do we vote to accept this report? Just raise your hand. I see its unanimous, sans Miki was not here. So, let the record show that.

And with this, we can take a break. I suggest we come at quarter of ten, 30-minute break as planned. We're running a little ahead, but I'm sure we'll spend it somewhere along the way.

DR. LANZA: And again, please consider prepaying for your lunch.

(Break)

Agenda Item: Response to Review

DR. LANZA: We're going to have a response to the review by Dr. Robert Heflich and Dr. Manjanatha. We'll then have a discussion and a statement by RADM Denise Hinton. So free to go. I remind you that we have 15 minutes allocated and so I'll try not to burn our little advantage here in this next review. Thank you. Please, Dr. Heflich.

DR. HEFLICH: First of all, I should introduce myself. I'm Bob Heflich. I'm the director of the Division of Genetic and Molecular Toxicology. And as you heard from Dr. Felter, our division's programs were reviewed in March in association with this open SAB meeting. And as part of that process, Dr. Felter and the Review Subcommittee produced a document with all the comments in it that she went over this morning. And we are asked to respond to that document in written format. And then the next step is to respond in oral manner at the next SAB meeting, which is right now. So, this is the Division's response to the comments that were made by the Review Subcommittee.

First of all, I'd like to thank the Committee for their work. And the chair and the co-chair in particular, they really kept us on schedule, and we had a very efficiently conducted meeting. At the time, I really enjoyed it. When I got the comments back, I realized it wasn't such a great performance in many ways. But I'll go over that in my responses.

So, as I just said, the co-chair with members of the SAB, Dr. Felter and Dr. Aschner, our subject matter experts who did I believe the bulk of the reviewing, where three individuals with a lot of genetic toxicology experience. David Eastmond from UC Riverside, who is a former EMGS, Environmental Mutagenesis and Genomics Society - it's a specialist society for genomic tox - is a former president in the early 2000s. And Ofelia Olivero from NIH is also a former president of EMGS. And the third person is Mark Fielden, who I don't know as well, but he has a lot of experience with biomarker development and he headed a genetic toxicology group at Amgen at the time. And I've heard he since moved on. But anyway, those who are three subject matter experts who conducted the review.

And before I get into the comments, I just wanted to say a few words about what our intention was in how we set up the agenda for the review and the emphasis

we placed and the review process. It was my sort of naive goal to engage as many people from the Division as possible and in the Subcommittee review. And the way I tried to do that, because we were limited in the time we had and the number of platform presentations we could actually make, was to encourage people to show posters at the meeting. So that's what we did. We had 17 posters presented at the meeting.

I've been at NCTR for over 40 years. So, I've been through this process in the past. And the comment is always made that we have to encourage interactions between the Review Committee and the Division being reviewed. And we really try to do that through both the posters, and time was left - as is true of this meeting - a specific amount of time is left after each presentation for questions and answers from the from the audience. So at least we tried to do that. Because this is another comment that's often made, we also didn't want to present a lot of stuff that we've already done and was kind of easy to present because we already had the slides and everything. But we wanted to maximize the number of talks we gave on new projects, even projects that were just concepts at the time. And just to get feedback from the Committee as to

whether or not they were good ideas or not, and whether we were going in the right direction of them.

So, we had a number of short form platform presentations. There were two longer platform presentations by Dr. Parsons on cancer driver mutations, which had a lot of new information associated with it and a lot of plans for the future. And also, from Vasily Dobrovolsky, who gave the update on what we're doing to gain regulatory acceptance for the Pig-A assay, because that's one of the things that we do that has direct relevance to FDA regulatory needs. And in fact, CDER has supported us to do a lot of that work over the years. So, we thought it was important to feature those two plus a lot of other shorter talks that were more developmental in nature and speculative.

And in doing this, we limited ourselves to three topic areas. Dr. Felter already went over these. What we do for FDA regulatory needs, and that's obviously our major mission, but more sort of proactive kind of areas where we're trying to determine how we can adopt some of the new genetic analysis techniques, the regulatory decision making, and use some of the new biological platforms in our particular area of expertise in genetic toxicology and inhalation toxicology.

So, in making these choices, we left a lot of things out. And these are two of them that were left out. That was mainly because they were either externally funded or they were mature or combination of the both. So we didn't say a lot about our work with the in vitro airway AOI tissue model, which is a big part of our research portfolio, and the application of dose response modeling to applying risk assessment methods to genetic toxicology, which is a big area of current interest in the wider genetic toxicology community these days.

So, our agenda is listed here, and you've heard some of this. This is sort of redundant to a certain extent. But I want to point out sort of the flow of the meeting. We started with almost an hour overview of the meeting that was given by Manju Manjanatha, who is Deputy Division Director. Manju isn't going to participate in this response, but he will give the Division overview. I think it's the last talk of today. So, if you're still around, you'll get to hear Manju. Then we had our Topic 1, supporting FDA regulatory needs. And Vasily, I said gave a longer talk, but myself and Rajan gave two five-minute sort of flash talks on two projects we were engaged in to help particular people in the FDA on making decisions.

And that was followed by a poster session. And unfortunately, the poster session was really overloaded with posters. I didn't realize that showing these posters sort of made the reviewers think that they had to review all the posters. And I guess I didn't realize that at the time. And the other problem was most of the posters were under one topic. So, one poor guy, David Eastmond, got to review the bulk of the posters and he complained about it to me later. But anyway, I'm sorry that happened, but that was a mistake. The second day was continuing with Topic 2 and 3, and a follow-up. So, it was two half days, total of about eight hours.

So, first of all, I said this before, I'll repeat it. We thank the Review Committee for all their hard work. They obviously went through a lot of the materials that we gave them and came up with some nice recommendations for us. And we, first of all, can appreciate the kind words that that they said about us. Dr. Felter read some of those this morning, so I won't repeat this. So, the rest of the talk is going to be responding to some of the comments. And what I've done here - these are all text slides, so they go fairly quickly - but what I've done here is I've highlighted the Committee's comments in yellow and our response - and this

is excerpted directly from our written response - are in italic bold font. So, should be easy.

The first was the issue of the posters. As noted elsewhere, it was not possible to thoroughly review and discuss the posters with their authors, given the limited amount of time available for positive viewing. I can only apologize for that. It was totally my fault that I set that up. Just throw this in, the next division being review, which is neurotoxicology is using the same format, but they're not giving as many posters. They learned from this experience. So, this will be a lot easier on the reviewers when they come into the review, that division.

This is more of a reminder, I think, than a criticism or a comment. Encourage NCTR to continue to keep division reviews focused on those projects for which input is specifically sought. And it is not possible to conduct a complete review of a division within one day, which is obviously true. And that was our intent in limiting the talks to - at least the platform talks - to topics that we were seeking advice on.

Now a couple of things leaked through, even though I did not intend to talk about them. But one of the subject matter experts was an expert in risk assessment. So, he made some comments specifically about the dose

response modeling we had done, that came up, I believe, in one or two of the posters. This is the comment about the questions and wanting more time for questions and interactions. And I agree this is the most important part of the review process is the interactions with the experts. Over the years, I've gained personally a lot from that interaction. We should try to do as much of it as possible.

And I just mentioned a couple of suggestions here that might facilitate that. You probably don't realize this sitting in front of this room, but it's really hard to hear anything in the back of the room and I think it would help everything if at least the people who were involved in the topic could sit at the table where they could interact directly with the subject matter experts and the people coming in to conduct the review. It just makes it a lot easier. And maybe two topics instead of three. I don't know. And we could have the posters directly after the topic, so there's a better connection between the two. Something to think about.

The Subcommittee was encouraged - this is about the benchmark dose business, the does response analysis - was encouraged to see that the benchmark dose analysis had been included in a number of the poster presentations.

However, there was considerable inconsistencies in the approaches used, different benchmark responses, et cetera, and the work appeared to be more of an afterthought and not an integral part of the studies. Now I single this out because this is a particular interest of mine. Unfortunately, this was not one of the areas we emphasized, so we didn't explain it very well as far as its significance and I think its future as far as genetic toxicology is concerned. But this is a mature area for DGMT.

It was actually started via a CRADA-funded vehicle in the 2000s by the preceding Division Director who preceded me, Martha Moore. And the application that was probably given in the posters, which is what is referred to as genotoxicity potency ranking, a procedure that was first described by a member of our division in a paper in 2016, and since has been adapted by other people. And unfortunately, I realized when I read this yesterday that these comments have a lot of citations in them. So, I made a sheet with all the references in it. So, if anybody's interested in any of these references, I can give you this information so you can find where this comes from.

I'm not sure if this is helpful for you, but it's interesting to me. The variability in benchmark responses or BMR, or critical effect size as it's sometimes called, picked up by the reviewer, stems from a lack of consensus on the most appropriate BMRs or CESs for determined relative genotoxic potency. This is a relatively new area for genetic toxicology, so the critical effect size for the various assays has not been rigorously established. So, what we did is we tried a bunch of them, and we essentially saw which worked best. And as I said, many researchers are sort of zeroing in on Benchmark 50 for genetic toxicology. Whereas for cancer, it's usually a BMD of 10, 10 percent over the background of 5 percent, sometimes. Or one standard deviation one time depending on what regulatory agency you're working with.

And I just want to point out that potency ranking was not used as an afterthought but was intrinsic to our conclusion that genetic toxicology dose response data coupled with BMD potency analysis could be used to rank order the toxicity related test agents. And this came out of a study funded by CTP on cigarette smoke extracts and that was published in 2018.

Now, the Subcommittee - and this is probably particularly Mark Fielden - had a lot of suggestions on our research using error-corrected NGS. And for those of you not familiar with this term, I'm sure you know about next-generation sequencing or NGS or massively parallel sequencing. It's very error prone. So, you can get every hundred bases, you'll probably get a mistake. But NGS overcomes this by doing the analysis over and over again. So, you get a consensus set of that. If you want to use the power of this platform for looking for rare events, you've got to sort of take several extra steps to be able to correct that error proneness and get the background down. And there are several ways that have been employed recently. They're sort of new and we're kind of evaluating them to see how well they all do.

This first comment is address what set of - the error-corrected NGS was used to analyze cancer driver mutations in this presentation made by Dr. Parsons. So, she had used other techniques previous to this, but was starting to adapt a particular form of error-corrected NGS, or EC-NGS - so address what set of driver mutations will be important to assess for specific tumors of interest and how these may behave in non-clinical models. The DGMT should continue to avail themselves to both non-

clinical rodent models and clinical samples to further test these hypotheses and improve our understanding of the translatability of rodent data.

Now, most of what Dr. Parsons presented at the meeting was human data, so that's where this comment comes from. But our response is, we fully agree with this. We fully agree with the Subcommittee's suggestion of developing panels of CDMs appropriate for different tumors and for human and animal models. In fact, this is the current focus of Dr. Parson's research, as described in a recent review paper published by Harris et al. Kelly Harris is a staff fellow working with Dr. Parsons on this problem. Since the review of the Parsons lab adapted their EC-NGS method that they've coined CarcSeq to the analysis of analogous, conserved hotspot codons in rat and mouse. So that's ongoing work.

More into the nuts and bolts of the EC-NGS. I think I sort of jumped the gun there. DGMT should consider developing experience with other error-corrected sequencing approaches as they become commercially available. This was actually underlined in the written comments. There is one commercial source of this. The evaluation of other commercial EC-NGS approaches is also underway. Dr. Parsons has contracted with TwinStrand

Biosciences, which is a startup in the state of Washington that Dr. Fielden is associated with, or was associated with, and he was probably particularly referring to. And this company feels this is important enough to develop their own panel or adapt the panel to their own methods for EC-NGS, similar to one she has used for evaluating normal human lung and breast tissues. And the idea that the sensitivity and specificity of the duplex sequencing technique that they use, employed by TwinStrand, will be compared with Dr. Parsons' CarcSeq method. So, we're sort of playing around with these different methods to see essentially which works best for our problems, our issues.

So the Subcommittee had some more suggestions related to the talks given by Dr. Revollo and Chen, who used Illumina sequencing in their projects, and they said that short read sequencing technology, they used it, and may require single cell cloning expansion, which is the method they employed. If you take a clone and let it expand so you've got a lot of DNA, what you essentially see is you can use more or less standard NGS techniques to look at the genotype of that clone. Now if that clone happens to come from a mutagenized cell you can let that cell expand and it does all the work as far as error-corrected expansion of its DNA. It's done biologically.

And then you can just sequence that. So that's a clonal comparison method of using this.

And you can look at the entire genome so you can see very rare events because you've got all that DNA to look at. So even if there's only one chance change per genome, you can see it using that kind of technique. So, they sort of criticize that, that DGMT should consider exploring methods that can be readily applied to a variety of tissues, models, or species, and can be incorporated into existing toxicology studies where single cell cloning is not readily feasible. Well, I totally agree with this. The comparative cloning sequencing was employed by Dr. Revollo and Chen in their project because it was judged to be appropriate for answering the research question.

DGMT is currently using at least three EC-NGS approaches in its research, some of which are applicable to the cells that are difficult to clone. Since the Subcommittee review - and this relates to the Illumina sequencer. The DGMT has contracted to purchase a PacBio sequencer to complement its Illumina NextSeq 500. We anticipate that the PacBio instrument may be useful for detecting large events that are not detected efficiently by Illumina sequencing. And I won't go into why that is, but the chemistry is different, and the analysis technique

is different in this particular sequencer. So, you can get longer reads more accurately.

They did not embrace alternative models. The evaluation of base editing in E. coli and germline mutations in C. elegans, which were the subject of Dr. Revollo and Dr. Chen's talks, may also be problematic from the perspective of human relevance. Therefore, the use of mammalian models should be prioritized as feasible. Well, this is a perfectly reasonable comment. But I want to defend alternative models for a minute. We agree that mammalian models, especially human models, are ultimately the most appropriate for making regulatory decisions about human risk. However, not all studies need to be or should be done in animal models.

Proof of concept studies, binning responses as done with Hazard ID, prioritizing research, et cetera, can readily, more readily be done in alternative models, very often, than they are with animal or doing them in humans. So, Dr. Revollo's studies were preliminary proof of concept studies that were done in collaboration with CBER for looking at off targeting sequence changes. But the bacterial system itself may be useful as a primary screen. That may be true, also.

The use of C. elegans in Dr. Chen's study is an attempt to devise an alternative model for a field. And this is germ cell mutation. That historically has been held back because of the difficulty involved in generating experimental data. This is especially true for female germ cell data. Worms may not prove to be an adequate substitute for humans, but they were judged to be worth considering in this case. So, this was sort of a preliminary look see kind of thing that I thought might be interesting.

To the reviewers, you might know C. elegans is hermaphrodite. It is both a male and a female. So, depending on the timing of when you treat, you can treat male germ cells or a female germ cell. So that makes it kind of a neat system for looking at germ cell mutation.

They had some suggestions about our in vitro airway tissue model. I happened to hear this through the den in the back of the room when Dr. Felter was giving her comments. One of the components that should not be forgotten is the extracellular matrix and it's about 300 interactive molecules and of critical importance for the end points in question are the infiltrating immune cells that on multiple occasions are the producers of oxidative

stress and consequent DNA damage. We acknowledge the importance of extracellular matrix to tissue function.

I didn't get a chance to explain this, but one of the features of the human airway model is that it makes its own ECM, and in fact we've used ECM modification as an endpoint for toxicity. And that's given in these two papers that I cite. We have considered augmenting our airway model with endothelial, immune, or other cell types. I might say this this particular model is somewhat unusual. It forms itself from stem cells and it forms itself into four different cell types, including goblet cells, ciliated cells, basal cells, and club cells. And it remained stable over a period of months. So, it has very unique properties as far as in vitro tissue model is concerned.

And while we recognize the fact that it doesn't have all the cellular functions in it, changing that by adding immune cells, kind of throw something in that's not under the control of this kind of nice balance. And it would not enable us to do the kinds of experiments we've been asked to do as far as our collaborators are concerned. And that's normally long-term or longitudinal treatments of these cultures to see what effect - repeat those studies, inhalation studies, have on these

endpoints, because we want to model the humans. And often it's a chronic exposure. So, we want to keep as close to chronic exposure as is possible. So, there is a tradeoff here. We could add other cell types, but it would cause us to have a model that may only have a week or two useable life in culture. So, there's that problem.

In our current model, immune functions have been inferred by measurements of cell signaling molecules in apical washes and basal media, which cytokines and chemokines are usually signaling molecules for the immune cells to sort of invade the tissue. We can pick up on that. So, going forward, we will consider - we have always been considering adding more to this model and may well do so in the future if the research question asks for it. This is something that we've thought about a lot, and it's a good question.

The Subcommittee has suggestions for in vitro testes model. It is advisable that the model - and this is the presentation that was made by Dayton Petibone. It was the last presentation. It is advisable that the model be developed to obtain sperm cells, that in turn will provide a venue to address a different set of toxicities associated with exposures and associate them with endpoints such as malformations, motility issues, and

others. Another very good suggestion, and it is a goal of our work. But the current technology does not allow full spermatogenesis to mature sperm. You get partial spermatogenesis.

So, one of the goals of our work with testicular models is to develop methods that will enable progression through all the germ cells stages from germ cells to mature sperm. We, along with other scientists working on these models, continue to make advancements in characterizing the testicular organoids while devising improved formulations and culture methods that support the spermatogonial and stem cell niche and promote in vitro spermatogenesis. So, this is another study that we're just getting into and there's a lot to be learned and a lot to do here, if we're going to go forward on this.

And this is sort of good advice that I want to point out. We encourage DGMT to revisit its strategic plans on a regular basis and to ensure that there are mechanisms in place to gauge productivity and success towards short-term and long-term goals. I think this is something we do in the Division and it's something that NCTR does through its protocol review process. So, it obviously is a good thing to keep in mind. Although you can think of scientists that have labored away in complete

isolation for 20 years on a problem and all of a sudden, they find this fantastic result that the world is waiting for and know about it.

Okay, biomarker qualification. The DGMT should consider how their experience in validating the Pia-A assay could be leveraged to help develop guidelines for how new genetic toxicology tools should be qualified for regulatory use. Additionally, DGMT is positioned to improve the qualification process of new biomarkers or models, which is currently quite lengthy. And I can agree with that, being involved in some of this, I always see the test guideline can take anywhere between 10 and 20 years to get finalized, at least in the world of genetic toxicology.

So, here's our response. We consider identifying and validating new genetic biomarkers and working toward their acceptance to be a major part of our mission. And in the future, I can foresee developing tests, if they prove reasonable, for involving EC-NGS for gene mutation analysis. That could revolutionize the field. The use of cancer driving mutations as a biomarker of cancer risk. This is something that people are interested in as an adjunct to the traditional rodent cancer bioassay and tests involving in vitro ALI models. And this is actually

already started to a certain extent to evaluate inhalation toxicants.

Dr. Parsons is a member of FDA's Biomarker Working Group and is on the Planning Committee for a Public Symposium on multi-endpoint biomarkers. Efforts aimed at furthering and standardizing FDA's regulatory use of biomarkers. Now this comment about speeding the conduct of FDA's biomarker qualification process, I feel a little uncomfortable about that we should be telling the product centers to speed it up. Because they're going to have to use these data for making their decisions and they're in the best position to know when something is ready to be used for evaluating a public health question. So, I tend to think that we can make suggestions, but I think that it's ultimately the decision of the product centers as to how they handle this.

The Subcommittee made some general suggestions for filling research gaps. It's important that DGMT consider the external landscape and the need to avoid reproducing what has already been developed elsewhere - they cite the human lung and testicular platforms - so as to focus Division resources where significant gaps in the field exist.

I would contend that these are two areas that were very strong in and impactful in. Well, one area where we're very impactful, and the second area is an under-researched area. But this is good general advice. We feel that we are leaders in research using the two platforms that are cited in this comment. Dr. Cao, who is in charge of our airway model research, is considered an expert in research in the in vitro airway model. And she receives many invitations to speak and prepare manuscripts on the model. Probably too many. The most extensive work conducted with this model was not discussed because virtually all of it is externally supported by CTP and/or NTP, and has become an important part of their, as well as our, research portfolios. So, the Committee may not have appreciated it.

As far as the testicular 3D organoid models, they are one of the least developed in vitro tissue models, largely due to the complexity involved in promoting spermatogenesis. And in fact, I know of no one else that is investigating mutation inductions, whether or not they are reasonable models for studying germ cell mutation induction. So, we have a feeling we have an opportunity to develop this in vitro platform and

determine to what extent germ cell mutations can be evaluated using an in vitro culture model.

And I just want to add this without going into detail, all these models have alternative applications besides genetic toxicology, obviously. And as far as this testicular model is concerned, one of them that's come up recently was via FDA Medical Countermeasures Programs. It turns out that Zika virus has an infection site in testicular tissue. This is of interest to them as far as the control of Zika virus infection. Dr. Petibone was invited to submit a proposal for using his in vitro culture to model in support of their programs.

Okay, career development. We should consider initiating or enhancing career development programs. And this says specifically for post-doctoral fellows. Obviously, a good suggestion.

This is what we do at NCTR, not only in our Division, but NCTR-wide. DGMT postdocs have face-to-face meetings at least twice a year. And this is a standard review practice that we have that for government employees, that we also include the postdocs in. So, we have time to discuss their career goals and options, because obviously that's the most important thing that the postdocs have on their mind.

I'd also like to say that under the guidance of the Deputy Director for Research at NCTR, Dan Acosta, who is now retired, he really developed an incredible program for postdocs and fellows in general, a wide range of opportunities for career development, including there is an organization at NCTR that has its own organization for postdocs that have regular scientific and social events. We have a NCTR Science Day that's sponsored by this organization. So, in the last couple of years, this has really blossomed as a real positive thing that goes on at NCTR. And I was very happy that someone took the initiative to do this. Fellows also had an opportunity to take a regulatory science certificate course that is taught at the University of Arkansas School for Public Health. And it's fully supported by NCTR financially, so they can just sign up for it. And a number of people have done that and taken advantage of that. They actually get some kind of a degree.

This is something I want to add about career development because it's something I miss. Until recently, the FDA had a Commissioner's Fellowship Program that combined courses in FDA regulatory issues with laboratory research in a two-year program. Unfortunately, this has sort of been phased out. My understanding of it. But it

was very successful in the last few years, as far as our Division is concerned, in developing potential staff scientists. And to cite two of them, Dr. Cao and Dr. Revollo, both of whom I mentioned, are graduates of that program and others have moved on to careers in FDA Product Centers and industry.

This is a problem that you may hear more about during the course of the SAB meeting - post-doctoral recruitment. We recommend that NCTR establish relationships and increase outreach recruiting efforts with graduate and other training programs to ensure a pipeline of eligible and qualified post-doctoral fellows and early career scientists, enhancing the diversity of the ORISE fellows will add originality, creativity, and innovation.

We agree that recruitment, especially post-doctoral recruitment, has become an issue in the US Federal government in general, with the rules that are in place, it's very difficult to have foreign nationals in as postdocs or even as a visiting scientist. Just the rules are set up such that discourage that. And because of that, we're trying to adapt to that. It's been something that's affected us a lot. We are trying to establish better connections with US-based institutions. Most of our newer

postdocs got their Ph.D., as far as ORISE postdocs, got their Ph.D. in US institutions, and that's almost by necessity. And it's obvious that using professionals, society recruitment services are also something that we have been doing and we will do more of in the future.

DGMT, if you've met us, you can appreciate that we're a diverse organization as it is. I'm not sure what effect these restrictions are going to have on the future, that diversity, but we can hope that it remains a diverse organization.

The Division should also consider succession plans for their staff to ensure continued career development and retainment. This is obviously something that's discussed NCTR-wide. It's something that I personally tried to do in the years that I've been a Division Director to promote people and convert people to government positions, when they are available, when the opportunity presents itself. And with the idea of developing new PIs, we have 35 members of our Division, more or less with postdocs and everyone, and about a dozen of them are PIs. I think that's a good ratio of PIs and an equal number of support scientists, and then a bunch of postdocs. So, I think that's a good mix, if our numbers are going to remain static. So, we have to constantly

think about who is going to be the next PI and who we're going to move up the chain.

One of the real advantages at NCTR - and I guess this is FDA-wide - is a Research Scientist Peer Review System, which gives an opportunity using a committee approach, peer review approach, to decide on people's grades and promotion. And this takes the decision out of the hands of one individual and gives it to a committee of experts, which is really something that I think is of benefit to the scientists that work at NCTR as far as increasing the fairness of the whole process. And succession plans for Division leadership are more or less in place and they're being discussed.

And again, I'd like to thank the work that the Subcommittee put in on looking at us and preparing a report. I open the floor to questions if anyone has any.

DR. LANZA: Thank you. That was very good. And I wanted to just make one comment and then we'll do discussion. I thought the report actually was very favorable and that the criticism was more like ways we could tweak it sans the procedural stuff. And I think that the comments that you've made in the implementation will just make it better. And I didn't want the group to have the misimpression, I think, that it was a bad report. I

think that report and your Division was very strong. I'll open it up to questions.

DR. FELTER: Thanks for that, Greg. I wanted to actually make the same comment. I do hope the report is taken in the way that it was meant, which was very favorable with a lot of strong comments in support of all the research that DGMT is doing. So, I wanted to start with that. I think sometimes something that you might have reflected as a criticism was a suggestion for future thinking that clearly you have to do what's feasible today and you have a goal for tomorrow. It doesn't mean that it's a criticism that it is not being done now.

I also wanted to say thank you for trying the posters and I'm glad to hear that the Division review that's going to happen at this meeting will continue to have posters. Maybe we had a few too many, a few more than we were able to handle in the time allotted, but the concept for why you chose to do that was appreciated. And I think the goals that you stated were apparent and I am glad that you did that. I do think it's important to give opportunities to the other researchers to have an opportunity to show some of their research, as well as emphasize the breadth of research that we don't get to see in just a few select presentations. So, I hope there is a

positive that comes away from that also. And not just a criticism.

DR. HEFLICH: I'm usually not very hesitant to pat myself on the back about this, but it was my understanding that we were supposed to comment on the suggestions and the comments for improvement. So, I concentrated on what I could find in the report and what I wanted to respond to in the report, in this presentation. But yes, I really do appreciate what you said. Whenever you get a job like this, you sort of inherit something that's already there. And I really think the Division is really a good group of people who are trying to push the field of genetic toxicology, which in many aspects is a mature field, into bigger and better applications for assessing human risk. And I really think there's a lot of opportunities out there that a Division like genetic toxicology can take advantage of.

Now, a lot of what we do for FDA is standard assays. We do the Ames Test, we do the micro-nucleus assay, and things like that because people need that kind of information. We're certainly capable of producing that. But there's also a lot of exciting things that we can do. And as a research organization, we try to partner up with

people in the product centers who also feel that way and sort of try to move the science forward.

DR. GANEY: So, with respect to the in vitro airway model and immune cells, I think you're actually doing the right thing by allowing that model to mature a bit before you start adding immune cells. But an alternative to adding cells would be to add soluble mediators that the cells would produce. And there are lots of challenges with that. Which ones? When do you add them? But I was wondering if that had been part of your thinking at all.

DR. HEFLICH: Yes. Actually, the cells produce mediators. The cell types that are in there are capable of producing a lot of signaling proteins that activate immune pathways and those kinds of things we can measure very readily. There are alternative models that utilize precision-cut lung slices, for instance, which only lasts for about a week or two in culture, but they have a complete immune complement that comes along with them.

So, in some ways, if you're looking for immunological effects, they might be a better model to use at least to try anyway and see what kinds of responses you get. There are few people who actually specialize on that kind of research, not only academically, and there's a CRO

in Gaithersburg that has an expert in that area also. It's a very interesting system.

DR. LANZA: I think we can take that one second point offline. Any more questions? Susan? Please identify.

DR. FELTER: I did want to offer just one other small clarification that it was related to the biomarker qualification process. And it was my understanding, again, I'm not the one who wrote that part of the report, but it was my understanding that when they were talking about leveraging the success that you've had with the Pig-A assay to help expedite other processes, it was not specific to FDA's qualification process, but more in general. So, for example, the Pig-A process was specifically talking about OECD's qualification process. So, I don't think it was intended to - I think it was meant more broadly.

DR. HEFLICH: OECD is nothing if not bureaucratic. And when you get 38 member countries together and they all have to agree on something, it's kind of like herding cats. As I said, it takes years to work through the process of consensus forming on some of these things. I'm not sure what I can do about that.

DR. RAMOS: Could you comment on the timeline for the Pig-A mutation assay? From start to where you are today, what is that time interval?

DR. HEFLICH: As far as the assay as a safety assay, a regulatory type assay, it was actually first described here at NCTR in 2008. And at the same time, a commercial lab in upstate New York, also published on it. We sort of knew they were working on it, but we weren't working together at the time.

The politics of the situation really were sort of primed for this area because there is an in vivo gene mutation assay called a transgenic rodent assay that is very capable assay, but it's really difficult to perform. And industry and all the regulatory agencies were also looking for something more practical that they could do using generic animals and just sort of integrated into standard toxicology testing, something they could do without huge amounts of expense using standalone kinds of assays. And that's one of the reasons OECD is interested in it. They're very interested in 3Rs type applications. So, the timing was right at that time.

So, it's been about 10 years. HESI, I should say, became involved in it. And they pushed the assay because they have both government and industry and

academic members to that. And they get very excited about it. And I'm the chair of that Working Group for Hessey, and we actually presented to the OECD a plan for a test guideline because that's the gold standard in genetic toxicology anyway of regulatory tests, is to have an OECD test guideline. A lot of countries will not use a test for making a regulatory decision, not FDA, but a lot of countries will not use a test unless it has an OECD test guideline associated with it. So, we made that as a goal. So, for four years we've been in the official pipeline for getting the test guideline in place.

And I can say I'm just putting together a revision of a review article. It's called a Detailed Review Paper and a Validation of the Assay. It's been reviewed by external - it's called PRP - Peer Review Panel for validation and it got very nice marks. So, I'm making a few tweaks to that. So that will go back to OECD and then it will go out to the WNT Review, which is actually all the countries. And at some point, Tracy Chen, the OECD contact for FDA will get that document and distribute it among the FDA Product Centers and they'll have a chance to comment on it. And then it will go back and hopefully WNT will approve it. And then we can go on and write a test

guideline, which I suspect would be the relatively easy part of it. And it should only take about a year.

DR. RAMOS: And so, in your experience, do you think those 10 years and the time that you've described is acceptable? Worse than expected, better than expected? How would you qualify that?

DR. HEFLICH: I think it was very relatively fast.

DR. RAMOS: Ten years?

DR. HEFLICH: Ten years is relatively fast from ground zero on an in vivo gene tox assay because from first publication to accepted guideline, that would be phenomenal to have happened. Unprecedented.

DR. RAMOS: That actually is a useful metric, I think, for us to keep in mind when we sort of examine what we are looking at.

DR. HEFLICH. Now, that's OECD. Now, FDA has its own biomarker qualification programs and they have committees that that view on things. And as far as the more molecular biomarkers like gene expression kinds of things and protein expression, they typically go through that process. And then once FDA accepts through this biomarker qualification process, my impression is that the industry can submit these kinds of data in the regulatory

packages and FDA will have some basis for making a decision on them because they have looked at the assay. That I think is a faster route to getting a test used by regulators, at least within the FDA.

DR. RAMOS: And a second question is, over the course of the review period, how many manuscripts were published by the group?

DR. HEFLICH: I'd say between what and what? 2000 - you mean as far as Pig-A is concerned?

DR. RAMOS: No. The total output of your Division.

DR. HEFLICH: Not 100. I'd say somewhere between 30 and 50 a year, maybe.

DR. RAMOS: Do you know, or do you not know? You haven't tabulated yourself?

DR. HEFLICH: Yes, I have, but I haven't really counted them. I mean, it's in our package that we provided. In fact, the Subcommittee complained about the number and the fact that it wasn't keyed into the particular topic area. So that's also something we can probably go into.

DR. RAMOS: So, they complained there were too many? Is that the complaint?

DR. HEFLICH: Well, too many, that's what we have. One thing I did draw the line on was people putting publications in their CVs, which really can balloon the size of these packages. So, I told everyone to just take the most recent 10 publications as examples and sort of give a number for everything else, because if you're around for 40 years, you publish a lot of papers.

DR. LANZA: Susan, last question.

DR. FELTER: So again, I don't believe that we were complaining that there were too many. It was an impressive number. I don't remember what it was, but it was given to us, the number of publications. The only suggestion that the Subcommittee had was it would have been easier for us to appreciate the impact of them if they were organized according to how they fit into the different themes that were being presented instead of being in chronological order. Because when they were in chronological order, it made it more difficult to see, oh, these 10 papers all fed into this project. We were impressed by the number of publications.

DR. HEFLICH: We also provided lists of projects and things like that. And I'm not sure how useful it was either, except to see that it was a big list that we had

projects. The individual line items may not have been that useful for the review.

DR. COSENZA: I just had one quick question for the Pig-A assay. Are there plans to add that to the ICH guidelines or any discussions on that yet or do you have to wait -

DR. HEFLICH: That's up to ICH. That's a separate body. And right now, it's being used for regulatory purposes by FDA. I can say it's in the ICH M7 Guidelines for Impurities, because very often you get gene tox positives as impurities with drugs because you have no control over that. You know, usually drugs that are genotoxic never make it to the review process for FDA. The API is genotoxic unless it's a cancer chemotherapeutic agent. So usually that's not an issue. But where it becomes an issue is the impurities that come along with the manufacturing process. And yes, the Pig-A assay is already part of the ICH M7 Guidelines, which is the impurities guidelines, specifically because it's the best alternative there is as far as generating test data. And this is test data - usually they have very small amounts of test compound and they want to integrate the tests together. If they're going to do an in vivo test, they want to do several tests all at once. So, this would be a

good way of getting gene tox data and also micro-nucleus data out of a single set of animals, along with general tox data, which is often the way it's done.

DR. LANZA: Thank you very much. Thank you for the questions, and I think I'll close the discussion on the response to the review from Dr. Heflich. And I want to introduce RADM Denise Hinton, who will provide a statement as the Chief Scientist.

Agenda Item: Statement from the Chief Scientist

RADM HINTON: Good morning, everyone. I'm entirely pleased to be here with you today as FDA's Chief Scientist and working with each of you. As we've already heard throughout the morning, NCTR's work and contributions are their footprints all over everywhere within the FDA Centers and often central to some of the FDA's top priorities, as you've seen this morning and will to continue to throughout the day.

In listening to today to NCTR's achievements over the past year, I think you will have to agree with me, NCTR has been on the move, literally and figuratively, and making remarkable contributions both within the Agency and with our domestic and international stakeholders. The amount of data that NCTR generates to support FDA decision

making is truly impressive, as is your leadership. Thanks to all of you.

On that note, I'd like to congratulate Bill Slikker, who was one of the recipients of the Commissioner's Special Citation Award for the 21st Century Cures Task Force on research specific to pregnant women and lactating women. I commend you and your group for outstanding contribution to developing an industry-wide inventory of initiatives for pregnant and lactating women, along with HHS recommendations for clinical trials. This is a precedent for us. Thank you for that.

Before going any further, I also want to acknowledge Donna Mendrick. She, together with FDA Center and OCS members of the Toxicology Working Group received the FDA Group Recognition Award for Developing and Promoting an FDA Roadmap. And this is to incorporate new predictive toxicology methods and methodologies into regulatory science. We had Dr. Susan Fitzpatrick and many of you around the table engaged in that, and we really commend your efforts in that. And we'll be doing more in that space as we move along. I want to thank you for your work. You have also chartered FDA's course for working with our many stakeholders at sister agencies and industry and academia, as well as the international level in this

pioneering area of science. And thank you also for the progress you and your Center colleagues and the Toxicology Working Group have already made. And I'll get back to those points in a minute.

I think we all recognize how critical NCTR has been to the development and evaluation of emerging toxicological methods and other new technologies that play a large role in FDA's regulatory decision making. As I've said so many times, NCTR holds a unique and foundational position at FDA because it is the only Center that supports all FDA offices and Product Centers with the essential toxicological research they need, to conduct their scientific activities. It is also the only Center that is situated within the Office of the Chief Scientists, AKA, OCS. This is no organization anomaly. It underscores the criticality of toxicological research for everything FDA does to advance regulatory science.

I was very pleased to join you recently at the October ribbon cutting ceremony for the grand opening of NCTR's Building 14. This brand new $26.8 million facility includes renovation of over 16,000 square feet of lab space and 10,000 square foot addition for new offices and new lab casework. As you've just heard, the new building will enable us to benefit from a broad array of research,

including biomarker identification and development, evaluation of the toxicity and inflammation produced by cigarette smoke, developing quantitative analysis tools for compounded or adulterated products, and developing tools to evaluate the mutagenicity of FDA regulatory products.

FDA will also benefit from significant upgrades to anti-terrorist buildings 53-A and B, which include 11 new labs and the replacement of an antiquated processing area essential to animal research. These are ambitious achievements, and I will say right now that my office has been, and will continue to be, fully committed to raising awareness of NCTR scientific research and its impact on our regulatory decision making. Supporting NCTR and its work to protect public health and advance innovative tools and approaches that are critical to FDA's predictive capability and our ability to predict risk and efficacy.

NCTR's research continues to be a regular feature of our monthly FDA Grand Rounds, the webcast that OCS launched back in 2016. The goal of the Grand Rounds has been raised for the visibility of FDA's research in the scientific community and describe how FDA is applying that research to its regulatory activities. This past July, NCTR's Dr. Amy Inselman presented some very exciting

research underway as part of FDA's Perinatal Center of Excellence. The Center was established by NCTR, as you heard, to coordinate agency-wide research that addresses special public health needs during the perinatal period. FDA regulated products given to newborns and infants or to pregnant mothers that haven't been studied extensively in such populations, leaving knowledge gaps about safety, efficacy, or potential toxicity of these products.

Knowledge gaps about raising awareness also exist about environmental exposure through foods and because infants consume more food per kilogram of body weight than any other age group, the potential is greater for dietary exposure to chemicals. The NCTR's Grand Rounds presentation highlighted Perinatal Health Center of Excellence funded project to investigate opioid-induced neural tube defects in a mouse mode to help clarify the link between maternal toxicity and embryo fetal development following opioid exposure. The results of this project may inform future label changes that can help pregnant women and healthcare practitioners make more informed decisions about the risk of opioid exposure during early development.

And also, with the Grand Rounds, we can't forget about moving forward with this past year, also September

Public Science Forum, also supported by OCS. NCTR was involved in every aspect of the Forum's planning and shaping its key topic areas, including tools to predict toxicity and efficacy of FDA regulated products in humans and in animals. I'm pleased to say that the event was a success FDA aimed for in reaching the scientific community with 74 percent of attendees from industry, academia, sister federal agencies, international governments, and other organizations. Yes, this is all show and tell and all about NCTR. I'm pleased to brag about them.

In September of this year, the Toxicology Working Group held its public meeting on FDA's Predictive Toxicology Roadmap to share with stakeholders FDA's continuing work to support implement the roadmap. This is a total of 521 academic, industry, and federal stakeholders that had attended. The group has published an annual report which is available on FDA's website on FDA activities that have advanced predictive toxicology. And you will hear more about today. Members of the Tox Working Group have continued to participate in, and lead interagency groups such as Tox 21, ICCVAM, to further our collaboration in advancing the goals of the roadmap.

The group's efforts also involve the formation of an In Vitro Systems Working Group, of which NCTR has a

leadership role in. NCTR has an important role in a number of FDA working groups, in addition to toxicology. I'm thinking especially of the efforts that we're supporting in OCS, like the groups working on in vitro systems, emerging sciences, and artificial intelligence to name just a few.

And as part of FDA's efforts to bring the latest innovative technology to its regulatory research scientists, my office is announcing a new webinar series on predictive in in vitro, in vivo, and in silico methods. Scientists will have the opportunity to present their new methods and methodologies, and this will be an ongoing series that will be advertised to FDA scientists exclusively. More to come on that. And NCTR will be directly involved in that as well.

One project I'm extremely interested in involves the Emerging Sciences Working Group. NCTR has been spearheading heading efforts to scan the horizon for future trends in science and technology that may affect products in our regulatory portfolios five to ten years down the road. This is a critical effort and I commend Donna Mendrick for leading those.

FDA will be able to prepare, by conducting in-house research and hiring staff with specific expertise.

For example, the group has met with US Government agencies, DDRA(?), NIEHS and EPA, as well as several regulatory agencies in other countries to learn about their efforts. They've also participated in the ICMRA, the International Coalition of Medicines Regulatory Authorities, strategic priority on innovation and in Workstream 1. This is analysis of global best practice in horizon(?) scanning.

The group has identified artificial intelligence as a significant tool and formed a new cross-agency group dedicated to its study and application in FDA's scientific activities. We appreciated your comments earlier on what more NCTR that we can also do in the face of AI The staff at OCS are very excited to be involved in AI's impact on regulatory issues and in identifying common need and ways to address them.

Finally, I'd like to recognize the vital role NCTR plays in promoting global harmonization and the standardization of regulatory science and its work with our international partners. Under Bill's leadership, NCTR established the Global Summit for Regulatory Science in 2011, which brings together leadership from nine countries in the European Union, each year, to focus on the regulatory science research. These partnerships, like the

Global Coalition for Regulatory Science Research, are leveraging global exchange, training, and collaborative research with toxicologists and other scientists worldwide to modernize the safety assessment of the products we regulate.

On the last note, I am reminded that this year's Global Summit, as Bill said, is taking place in exotic Bethesda, Maryland. So, this is September 28th through 30th, 2020. And it's on emerging technologies and our application to regulatory science. So, I hope very much that we will see you there and that you sign up early. I look forward to continuing to engage with you across the week and discussing the many things that NCTR is engaged with and the representation of the work that they do in working with each Center in ORA. And now I want to turn the podium over to the Centers to kind of showcase our engagement and the collaborations that we do have. Thank you.

Agenda Item: FDA Center Perspectives

DR. LANZA: Thank you very much for that statement. And with that, we'll start with the FDA Centers, and initially, the Center for Biologics. And this is Carolyn Wilson and she'll speak for a few minutes. We're trying to make these 20 minutes each. Thank you.

Agenda Item: Center for Biologics Evaluation and Research

DR. WILSON: Hopefully, mine will be a little faster. I'll try to go fast. So, I want to thank the Chair and Dr. Slikker and Mendrick for the opportunity to present the Center perspectives on the first day of this meeting. This is a new format, I realize, and I hope that it's informative to the SAB in your review as you go through the next day and a half. I also wanted to take a moment to introduce Dr. Braunstein, who is here with me as a colleague who's learning more about NCTR and the programs that we're interfacing as collaborative projects. So just if you haven't had a chance to meet her, she's here.

So, the products regulated by CBER, we have a variety of diverse biologics, blood and blood components, blood derivatives, vaccines are sort of the most common bread and butter things that we've been doing for decades, actually over 100 years. But also, you may not know as we also regulate allergenics as both for diagnosis and treatment of allergies. That actually represents over 1,200 different allergenic extracts that you can imagine the complexity of that product group. It's one word, but it's big. We also regulate certain devices, the exciting

new areas of gene therapies, cell therapies, certain human tissues, live biotherapeutic products, which you've heard me talk about in the past that include things like fecal microbiota transplantation, but also things like bacteriophage therapies, which are promising new approaches to address some of the problems associated with antimicrobial resistance. And then xenotransplantation is another area.

Because of the complexity of the products we regulate, the fact that most of them cannot be terminally sterilized, as you can imagine, the source materials that they're derived from we feel as absolutely critical for our regulatory mission to have a very robust and active research program. We organize our research around four major goals. The first around the technology, reagents, and standards to inform and improve chemistry, manufacturing, and control. So, in other words, product manufacturer. The second is developing and accessing non-clinical models and methods predictive of clinical performance with respect to toxicity and effectiveness. The third is around clinical evaluation, pre and post licensure, using a variety of big data, innovative designs, and statistical analytical and modeling approaches. And finally, but very importantly, as you can

imagine in this space, is preparing for future regulatory and public health challenges.

I've organized my talk around the goals that the collaborations that we have with NCTR address and they really address Goals 1 and 2. Primarily, as you can imagine, with NCTR's focus on toxicology that most the collaborations are supporting Goal 2.

So, within CBER we have a wide array of scientific expertise. We have expertise in a number of useful applied technologies for analyzing the products that we regulate, such as high-resolution NMR, mass spectrometry, flow cytometry, microarray, and high throughput or next-generation sequencing and the related bioinformatics and IT infrastructure to support that. As you would imagine with the products, we regulate a wide variety of microbiology expertise, biochemistry, molecular cell, and developmental biology. And more recently, we now have also expertise in tissue engineering and microphysiologic systems, epidemiology, biostatistics, and bioinformatics are additional bread and butter for us.

The facility that we have, we're on the White Oak campus. We moved there a little over five years ago from NIH and we designed it so that we would have expanded space to support a number of core technologies. I'm not

going to read the list for you, but it also has provided a state-of-the-art vivarium to provide additional capabilities in terms of imaging, BSL 2 and BSL 3 procedures and transgenic derivation.

On the left is a graph that demonstrates the breakdown of collaboration that we have. A question was asked earlier about, how do you leverage external partners. And as you can see, we have quite a variety of external partners that we collaborate with in a variety of means. But I do want to highlight for today's purposes the collaborations that we have with non-CBER FDA scientists. We have 33 ongoing collaborations. Nine of these are with NCTR scientists. And I've broken them down for the purpose of today's presentation and to those where CBER is supporting the expertise here at NCTR to support our needs. But there are also other collaborations where there's CBER expertise that is adding value to some of the work that NCTR has ongoing. So, I'll structure it that way.

So, regarding Goal 1, which is the CMC or Chemistry Manufacturing Controls, we have one project that you heard about from Dr. Heflich regarding detecting off-target mutations of gene editing and this is ongoing. Last year when I presented, it was really just getting started.

And this is a collaboration between doctors Revollo here at NCTR and Dr. Ye at CBER. And it's really important for us. As you can imagine, we regulate the use of genome editing in so far as it's being applied as a therapeutic product to treat human disease. And as was mentioned, the off-target effects are a very important issue that we need to address. And so, the experience that you heard about from Dr. Heflich in genotoxicity and next-gen sequencing has provided some additional richness to a study that we're doing to help address a significant regulatory challenge in this area.

A second goal that I mentioned is the non-clinical goal. And this is, as I mentioned, the biggest area. So, we have four projects here and I will be going through the first three in a little bit more detail in the next few slides and then not talking further about the norovirus diversity project because that's actually completed. It was published this year. It was very important, and enlightening study that demonstrated that in this case, canine noroviruses are sufficiently genetically distinct from human noroviruses that it's unlikely to jump species. So that's somewhat reassuring. We have enough of a problem with the human noroviruses

that are circulating. So, let me just dive deep into the next few projects.

So, the first is around Bordetella, pertussis and adhesion and pathogenesis. And this is taking advantage of the airway epithelial lung interface model that you heard a few moments ago from Dr. Heflich. And so, we're collaborating with a number of investigators in his group. And this is being led in CBER by Tod Merkel and Kelsey Gregg. Bordetella pertussis, as you may know, you probably think, oh, that's a disease that we're all vaccinated against with the DPT vaccine that you get as infants and children and we don't need to worry about it. But in fact, actually, since the introduction of the acellular pertussis vaccine in the 90s, there has been a slowly increasing resurgence of pertussis incidence, as many as 15,000 cases a year in the last several years. So, we realize that there is a need to develop a third-generation vaccine that doesn't have some of the adverse effects of the first wholesale pertussis vaccine but helps to have a more effective protection than the current acellular pertussis. So Tod Merkel is taking advantage again of the expertise here to develop this preclinical model that may allow for an improved understanding of pathogenesis, as well as addressing issues relating to

vaccine efficacy and hopefully provide mechanistic insights to help support regulatory review and as well as preclinical assessments of other respiratory pathogens if it proves out to be a good model.

The second area is a metabolic analysis on fecal samples, and this is a maturation of a project that I've talked about before with fecal microbiota transplantation. And Paul Carlson is the collaborator here and taking advantage of the extensive metabolomic expertise of Jinchun Sun. What Dr. Carlson has done is he has actually an MR1 which is mate cells a type of T cell knockout mice that has been shown to be resistant to Clostridium difficile infection. And actually, it's been shown that by transfer of the microbiota that that resistant phenotype goes with the fecal microbiota.

And so, the question came up in the review of this manuscript that he had submitted as to whether or not this could be residual differences in the cefaperazonein the mice because of differences in how it was metabolized. And so, Dr. Sun was able to very quickly show that there was no significant difference in the amount of residual antibiotic in the wild type and knockout mice. And now this paper has been published. So, this was a very successful collaboration and they're continuing to build

on it now with really combining their two expertise of the metagenomic analysis that's being done at CBER with the metabolomic analysis at NCTR to help identify other targets that might be worth pursuing. And I won't go into the details there, but this is a really active and exciting new area of research.

The other also involves Dr. Sun's expertise in collaboration with Dr. Akkoyunlu at CBER, where he's looking at using lipodomics to analyze macrophage incubated with sera. And the issue here is that there are certain polysaccharide conjugate vaccines against bacterial pathogens where neonates are known to be particularly unresponsive in terms of having an effective vaccine response. And so, this is an exploratory project to look at whether or not lipids may have anything to do with this non-responsiveness, and it may provide some new mechanistic insights to help inform development of effective vaccines in the future.

So, this last group are those collaborations where NCTR is taking advantage of some expertise within CBER. The first two are involving microfluidic systems, one on the mouse spermatogenesis and the second on a human placental barrier. And as I mentioned, we've started a new program on microphysiologic systems and it's really thanks

to the Working Group that you heard about from RADM Hinton that is on microphysiological systems that has allowed for a more rich interaction in the FDA of the experts in this field. And because of that, the expertise that we bring is in microfluidics. And so, we're enriching the development of these two models by collaborating with investigators down here.

The second is actually really a CDER collaboration with Oncology Center of Excellence looking at new biomarkers of doxorubicin-induced cardiotoxicity. As you can imagine, that really doesn't have anything to do with us. But there may be a need for a particular assay called Proximity Ligation Assay, which allows detection of protein/protein interactions and we have specific expertise in that area that may be applied there.

And then the last is, again, an area that's important to us, but we're not directly involved in terms of a laboratory component around whole genome sequencing and proteomics to look at markers associated with biofilm formation and the host specificity in methicillin resistant staph aureus.

So, what about potential future opportunities? And I think this air liquid interface model where we are already collaborating has some great potential to be

expanded. We realize that NCTR now is developing expertise with this VITROCELL Cloud, which allows this very precise manipulation of pressure, humidity, and droplet size and can be expanded to potentially allow examination of a variety of different lung related concerns from our perspective.

So, as I mentioned, we regulate all of the allergenics, and so microparticles of certain allergens such as pollens and house mite would be really interesting to study in this kind of model. We obviously are very interested in a number of respiratory viruses such as influenza, respiratory interstitial virus and so on. And we're also interested in vaccines that are looking at the mucosal immune response in the lung with and without particular adjuvants and the impact that has on this air liquid interface. So, we think that there is a lot of opportunities to expand some of the collaborations here and leverage NCTR's capabilities.

So, in summary, again, we like to think of ourselves as doing a good job in leveraging the expertise here down at NCTR to develop methods and approaches to support evaluation of our specific regulated products. Some new areas in FY19 are the lipodomics and metabolomics. And we've also expanded some collaborations

where NCTR is leveraging our capability in the microfluidics arena.

I think the challenges are pretty similar to actually what I listed here last year. I didn't change as much. Really the fast pace of scientific innovation and how to develop appropriate tools to evaluate the products. Identifying synergistic opportunities that address the regulatory and public health priorities. This is a challenge given the geographic distance. I think it always works best when it's a grassroots collaboration because they really identify their scientific needs in a better way than we can do it at an institutional level. But I think that to the extent that we can help promote that, that would be great. And then funding timelines and communication are just challenges again because of the environment that we live in.

So that's it for my presentation. I'm happy to answer any questions if time permits or maybe you're saving them until the end.

DR. LANZA: No, we have time for one or two questions.

DR. WILSON: Okay, thank you.

DR. LANZA: Thank you.

Agenda Item: Center for Drug Evaluation and Research

DR. LANZA: And the next speaker will be for the Center for Drug Evaluation and Research, this is Dr. Lal-nag.

DR. LAL-NAG: Great. Thank you. Good morning. And I'd like to begin by extending a warm thanks to Dr. Mendrick and Dr. Slikker for giving CDER the opportunity to talk to you a little bit today about our research program, our vision for standing up a research management system, and then delve a little bit into the importance of collaborative research with NCTR and other organizations. That's sort of been the theme of this morning. And I'd like to really reiterate again, and you'll see this in the following slides, that collaborative research is really the way to sort of move the needle forward, whether you're talking about translational research or niches in regulatory science research where we can really make an impact.

So, with that, I'd like to start by giving you an overview of CDER's research goals and objectives. Talk a little bit about the CDER Research Governance Council that was put together in March 2017 at the behest of Dr. Woodcock. Talk a little bit about our strategic plan and

how we've sort of outlined where we would like to go with research over the next five years and then move the focus a little bit towards scientific collaboration and CDER's Intramural Funding Program, which feeds directly into our interactions with NCTR. And then welcome the opportunity for NCTR to serve on CDER's Science Prioritization Review Committee, where we can really use the subject matter expertise to develop a continuum of a platform, if you will, for physiologically relevant models for pharmacological and toxicity testing.

So, with that, I'll start by telling you a little bit about the research loop at CDER. So, over the next five years, our vision really has been to stand up an actual research group within CDER that oversees all of CDER's research activities. Now, to put that in perspective, as you can imagine - and Carolyn has also sort of alluded to this with CBER - CDER's research activities expand a very - it's a very broad repertoire of research activities. So, to put all this together and to develop a vision for CDER research, if you will, is a little bit more complicated than one would imagine. So, I'm not going to go into or belabor all of the details but I'd like to focus on the four major areas that we've identified, which are the research drivers, the budget,

how our public health impact in integrating research within different entities. And then, of course, the eventual public health impact that we have.

So, in terms of our research drivers, we have the Congressional Mandates. And the reason I mentioned the Congressional Mandates is because of the Research Governance Council and how we came into being in terms of managing CDER research and the accountability for CDER research, but also our public health mission, public health crises, emerging science needs - that RADM Hinton spoke about - horizon scanning in scientific gaps. And what we're really trying to do, as I mentioned with the Research Governance Council, is focus in on these research gaps, not only within our internal research, but also with our collaborative research projects that we have with NCTR and others to reduce redundancy in research and increase complementarity in the research that we're doing as an Agency.

As you can imagine, in terms of implementing research, while the budget is very important, what is very, very important is the kinds of collaborations that we have. And when I talk more about the CDER/NCTR collaborations and Carolyn's talked about the breadth of CBER's collaborations with NCTR, this will become even

more apparent. If we're all able to work together in the pre-competitive space to identify specific questions - and I think Dr. Slikker actually threw out some very important questions for us to consider as a community in regulatory science research - I think we can really enhance the quality of research that we're doing, not only as a Center, but as an Agency.

In terms of public health impact, we put out policies and guidances. We have data registries, but with sort of non-nontraditional impact that we could be having that we have to work more on is in the space of joint fellowships, co-leading conferences with academia and industry. Again, this brings us to a point that was made earlier this morning about education and training. And I think that that pertains not only to scientific research personnel, but also to industry. So that the kind of data that we expect to see coming through the FDA in the next five to ten years is something that people have actually, developed as a group, have developed a set of standards that we can all adhere to as a community.

And then again, like I mentioned, over the next five years, CDER would really like to focus on serving the needs of unmet populations. So, with that in mind, I will go into CDER's Research Governance Council. And the

Research Governance Council, as I mentioned, was put together in March 2017 as a response to ORA report. We were put together to respond to the community with more accountability about where our research money was going and also to basically increase transparency and facilitate collaborations, not only internally but as externally as well.

So, the Research Governance Council, the vision for it is to be the benchmark for the governance of mission driven research for CDER. Our mission is to enhance CDER's research capabilities and its impact by fostering an awareness of, and optimizing, regulatory research activities. And I cannot stress this enough because given the flavor of research that we see within CDER, optimizing regulatory research investments is extremely important to us. Our goal is to establish CDER as a scientific leader and partner. And our strategic plan is a regulatory roadmap of how to get there. And I'll go into the main strategic focus areas of the strategic plan that we have for the next four years. We really hope to identify opportunities for engagement between regulatory and translational science research with NCTR and our sister Centers.

So very briefly, this is the structure of the CDER RGC. We have a Research Governance Council that looks through all - that manages not necessarily the research portfolios, but the bigger 50,000-foot view of where CDER should be focusing its research. We have a Research Tracking and Evaluation Committee, a Communications Committee, and a CDER Research Operations Committee that again manages the three sort of stalwart foundations that support what we call the CDER Research Program.

The CDER research goals and objectives were based and put together by the CDER Research Governance Council to form a framework that encompassed all of CDER's research activities. And this again was put together when the Council was formed in March 2017. The idea really was, is that it would serve as an anchor for all of CDER's research related activities, thereby enabling CDER to identify, organize, and summarize all of its research related activities as they pertained to each goal and objective. And the idea here again is to have better transparency in the way that CDER was investing its research money.

So CDER research goals are - we have five research goals and each research goal has six research objectives. The idea really again here is to make the

research goals and objectives as all-encompassing as possible so that everyone is able to identify where their research falls. And as a Center, we are able to identify where we're focusing our research and where we should be sort of redirecting focus for in terms of meeting regulatory science research needs.

The RGC Strategic Plan is a roadmap that has four major cornerstones to optimize regulatory science research, to influence regulatory science research, to serve the regulatory science research community, and to really foster an atmosphere of engagement and collaboration. And to do this at CDER's Executive Board for the Oversight of Research, the RGC governs research activities as a recommending body. So, one very important point to make here is that we do not make financial decisions. That is still made at the level of the Center and the offices. But we do make recommendations on where we should be focusing our research money after identifying gaps and facilitating collaborations to meet those gaps.

We influence engagement in CDER research activities. We've developed a panel of stakeholders and we actively interact with our stakeholders intuitively on a continuum to make sure that we're not missing anything that we should be engaging in. We are developing an online

hub that will be live in January 2020, that will be accessible to all our sister Centers as well. Again, it's meant to be sort of like a research gate, a source for everyone's ideas where you can actually partner and collaborate with things that may not necessarily be prioritized by budget, but prioritized by research focus or research need, if you will.

And again, the RGC really does aim to develop a community of trained scientists that can move up the ladder and sort of expand the scope of regulatory science research. In terms of CDER's Intramural Research Program, and this is where our interaction with NCTR is at its best, and NCTR really helps out in identifying niches that CDER could be actively contributing to. So the CDER Intramural Funding Programs that we have - and we have three of them, the Critical Path, the RSR, and the SRIG, they serve to basically identify gaps in CDER research and encourages collaborative proposals to basically fill these gaps that are not part of our regular research budget from an office-level perspective.

The CDER Intramural Funding Programs are the CEDR Critical Path, the CDER RSR and the CDER SRIG. The focus for each of the programs is slightly different. What we really want to do with the three different programs

that we have is to find a niche between translational science and regulatory science research, where the research needs can be developed and met for both the translational science research community as well as the regulatory science research community. Because at the end of the day, your patient isn't only the end user, it's your patient that has to be kept in perspective throughout the research process. And I do believe that having collaborative research agreements or collaborative research projects from the different perspectives can actually meet that need.

As I mentioned to you, we are three CDER Intramural Funding Programs, the Critical Path, the RSR, and the SRIG, which have a different focus. The CDER Critical Path is focused on innovative, cutting edge, emerging technology research, whereas the RSR and the SRIG are more safety related. And that's where we see a lot of collaboration with NCTR PIs.

In terms of interacting with NCTR PIs, traditionally we have, and we continue to do so, we accept concept papers and proposals on a rolling basis from NCTR throughout the year. And the Science Prioritization and Research Committee, which is housed within CDER, reviews this by interacting with a panel of subject matter experts

from across CDER. And in the interest of time, I'm not going to go through the whole process, but basically in terms of CDER inter-Center projects that are run out of the Office of the Commissioner, we are actually trying to trim our timeline so that we can get feedback back to NCTR PIs in a much more timely manner so that we are able to get these projects funded in a manner that is acceptable to PIs, so that everyone's not rushing to spend a million dollars over the course of one month before the budget year ends, because that is not really the best way to do science.

In terms of the NCTR Concept Paper and Protocol Reviews, as I mentioned to you previously, we have developed an automated system for all of NCTR's concept papers and proposals to go into. So that over time we have a system of referral and build up a bank of subject matter experts so that it facilitates greater collaboration between NCTR and CDER. Again, most concept papers and proposals come through the Science Prioritization and Review Committee, which is housed at CDER, and we send feedback back NCTR PIs. These are for the non-funded projects. This is just a scientific peer review.

Over time, there's been a tremendous impact of NCTR/CDER collaborations on review tools and projects.

From an informatics perspective, FDALabel as well as the Smart Template System were extremely beneficial and were developed through collaboration between NCTR and CDER PIs. And I'm only highlighting two toxicity studies. There are many. We have 31 active projects with NCTR right now. But in terms of the amount of focus that the opiates are getting at CDER right now, one of our collaborations led to a much better understanding of opioid exposure and effect on the developing fetal brain and nervous system. And the other one led to a much more comprehensive characterization of an induced iPSC cardiomyocyte system.

Again, this is just a snapshot of the collaborations that we have with NCTR or have had with NCTR over the over the 2018 to 2019 period. These are 31 projects by Division, and I have just taken one particular Division out again to give you a flavor of the kinds of collaborations that you have. And as you can see with the Bioinformatics Division, the different sort of collaborations that we have ranged from developing support systems to actual research predictive tools that can inform on toxicity studies. So, this is sort of the model that we want to develop for all of the different divisions. So that, again, as I said, you develop a

continuum of physiologically relevant models that you can use to study any regulatory research question.

In terms of the expertise exchange, again, I have just highlighted one project from each of the Divisions, but you can see that we've got a repertoire of different projects spanning BBD models from biochem tox to drug-induced liver injury from the Bioinformatics Division. And then we've been talking about the Pig-A assay all morning. We've got non-clinical modeling and risk assessment as well as the assessment of gaseous anesthetics in the developing non-human primates.

So again, whether you're talking about with lab research or in silico modeling, we are spanning the repertoire with our collaborations with the NCTR and really welcome much more of the same in the future.

Looking ahead, as I mentioned, the SPaRC, which is the Science Prioritization and Review Committee for CDER, would really welcome subject matter expertise and interaction with NCTR on this peer review committee for Center and Agency-wide proposals that come through to us. It affords an opportunity for NCTR to offer an alternative perspective as part of the gap analysis for our regulatory science research needs as they stand today. And it also affords NCTR a platform to showcase their research efforts

so that there's much more visibility of NCTR research to our CDER PIs.

And again, like I said, we really do welcome NCTR participation with CDER research projects and in our CDER research structure so as to ensure greater collaboration with the harmonization of multiple platforms. And with that, I'm happy to take any questions you might have.

DR. LANZA: Thank you very much. Do we have a brief question? A minute or two?

DR. COSENZA: Is there something preventing - on your last slide you said you would benefit from NCTR participating in this review. Is there something preventing that or -

DR. LAL-NAG: No, it's one of those things that has never been done. So, there's nothing in place that would prevent it. It's just something that sort of has to be facilitated.

DR. SAUER: John-Michael Sauer, Critical Path Institute. So, I have read through a couple press releases that you guys are forming. The Office of Drug Evaluation Sciences within OND. How is that going to affect this model?

DR. LAL-NAG: So that's an excellent question. So, as you know, we've gone through a reorganization and with OND and the Office of Clinical Pharmacology with OTS and OPQ being primarily affected. Again, since the flavor of the research and the drivers for research questions within these offices are so different, we have this research structure sitting on top of all of that so that we can sort of collate all of the different research that we see and direct people to address those gaps.

DR. LANZA: Thank you very much.

RADM HINTON: This is just a comment, actually. RADM Hinton, OCS. What I wanted to say is in facilitating that engagement, I think part of this meeting is helping to do that because we make plenty of connections throughout the years with each of the Centers and people within the Centers. But sometimes you may not always know, just because the Center is so large and there are so many ongoing efforts, what Committees or what Work Groups that people can engage in. I think from the Office of the Chief Scientist, that's something that we try to help facilitate and organize and structure from a cross-cutting look. And we do that through the work of our Senior Science Council, which are representatives from each of the Centers, including our ORA. But having discussions like this, being

able to come to meetings like this, it's kind of highlights the areas where there may be gaps and also opportunities for further engagement. So, I just say we stand ready to engage with NCTR and then also with OCS as a whole. So, thank you for that.

DR. LAL-NAG: Thank you. And OCS has actually been wonderful in sort of helping us set this up. So, thank you.

RADM HINTON: Thank you.

DR. LAL-NAG: Thank you.

DR. LANZA: Thank you very much.

Agenda Item: Center for Devices and Radiological Health

DR. LANZA: - is Dr. Margerrison. And this is the Center for Devices and Radiological Health.

DR. MARGERRISON: Thank you and good morning, everybody. I would like to take just a couple of minutes to take a slightly different tact from my predecessors. First of all, I apologize. I have no pictures on my slides. This to keep you paying attention before lunch. And I'd like to thank Dr. Mendrick for putting me on before lunch and not afterwards. So that's all good news.

CDRH, I think last year when I spoke late in March, I went through some of our Reg Science priorities.

So, I want to take a little bit of a different tact this morning and explain a little bit more about the program structure that we've put in place for our reg research within CDRH. And I want to give you a few examples of some of the challenges that we're facing as a Center and some of the things that we've done that we think are quite innovative. And I really want to put that out because we're to some extent reinventing ourselves from a reg research perspective at CDRH, and we can be a little bit of a different Center.

So, first of all, a little bit about the background. The Regulatory Mandate for us covers, as you would expect, medical devices. We also cover all of the in vitro diagnostics. Of course, we have a lot of crossover with our other colleagues in CDER, for example, for companion diagnostics. And we also have the regulatory responsibility for radiation emitting products. So that covers some consumer products as well, which I'm not going to talk about this morning. But it doesn't cover things like cell phone towers and the like, which are more infrastructure.

Regulatory Mandate covers a pre-market regulatory authority for all of these devices and diagnostics. We're also responsible for the manufacturing

facilities and the quality systems within those manufacturing facilities. And of course, post-market safety. And I think I threw some of these numbers out in March when I last spoke with you. But we have 190,000 types of product that we regulate. In terms of individual products, we're actually getting close to three quarters of a million right now. And I think I have to say my usual joke. There's 1,900 of us, which is about a third the size of Caesars Palace.

So, our mandate, many people ask what is a medical device? We quite often say that if you walk into a hospital, the first 100 things you see will be a medical device. This is our legal definition and it's really a lot of words that say it's a thing that is not chemical. So, if it has a pharmacological action or a chemical action or it's metabolized into something that has a chemical or a pharmacological action, then that's not ours. But pretty much everything else is. So, it really does cover a wide range of things from MRIs to CTs, and of course, all the other things that are coming towards us.

Now from our perspective - and I want to talk a little bit more about this on the next slide - we are very often the recipient of technology that's come from a different industry and then starts flowing into the

medical industry. Great examples of that are things like AR/VR that I'll talk about in a bit. 5G is going to have a massive implication on a lot of what we deal with. And what I want to do is spend a few minutes, really the bulk of what I want to talk about to describe what our research interests are before I talk about how we want to look at the future.

We have recently reorganized ourselves like happens quite a lot at FDA. This was not one that many of you may have read that the whole Center reorganized, and we now have a super Office of Product Evaluation and Quality. I deliberately kept my organization out of that, but we have still reorganized along different lines so that we can focus more not on, to be blunt, what degrees people did in university, but actually what our product lines are and what the therapeutic aims are of these products.

So I'm actually going to go through these one by one and just give you a little example of some of the things that we're dealing with and that we find interesting, because I think it's a little different from a lot of the other things that you hear about from my colleagues. AR/VR or augmented and virtual reality, classic example of something that's come obviously from

the gaming industry and is now very much a real thing in medical devices in our space. It's used a lot for not true diagnosis at this point. And one of the interesting things for us is where does it cross that boundary from something that aids a physician to something that actually is making a diagnostic assessment that then will be used for a clinical assessment. That then brings it into our world. That's one aspect of AR/VR that we're finding very interesting.

But it actually is broader than that for us as well because the image quality that a physician may be looking at is of utmost importance in the functionality of a device. And at the end of the day, our legal mandate is that we have to have to have reasonable assurance of safety and effectiveness. So, if an AR/VR screen or goggles are inaccurate or the color representation is wrong, that then device is not fit for purpose and we have to somehow make allowances for that. So, we have quite a large program actually understanding color and how monitors work and things like that. That will be very, very useful to this program.

Another angle that we're dealing with is we fully expect that in the future, the major interaction between a physician or a nurse or some other healthcare

professional and a medical device, a large capital equipment medical device, is going to be through some sort of virtual or augmented reality. We know that's coming. That introduces enormous regulatory science questions for us that we are actually going to be having a public workshop in the spring because we don't even know what the questions are yet, let alone how to answer them. So that will be a very, very interesting meeting and that will be a public meeting. So, I'd encourage everyone to try and be part of that.

Moving on to artificial intelligence, machine learning, that is clearly an area that affects all of FDA and all of our lives and is enormously important to everybody. For us, it's two aspects. And again, we're very different from the other Centers, I think, in this. There are many, many applications for AI machine learning in what I rather cynically called infrastructure. It can make our processes better. It can make our processes more efficient. We've actually been dealing with AI for about a decade within CDRH and we've already cleared two devices that actually have AI built into them. They're both diagnostics devices. One was for diabetic retinopathy and another was for Colles' fractures. But they're actually making those assessments through AI at the moment.

We recently published a white paper on how we think adaptive algorithms are going to affect a lot of our business. Again, to try and stimulate this discussion about what's going on out there. We know that algorithms that run a lot of the devices that we deal with on a day to day basis are going to change in the future. At the moment, we're anticipating, at least for the first generation, that the manufacturers will do a sort of a bulk update on all of those devices. But at some stage in the not very distant future, individual devices are going to be updating their algorithms on a day to day basis based on what they've learned that day. So, for us, that introduces really interesting questions. Do you need to revalidate that software before it's put into use? How do you do that? How do you monitor that it's safe and effective? Again, this is something that we're trying to stimulate a public debate about so that we can keep ahead of this. Very, very difficult for us to do this.

I think someone mentioned earlier the difficulty of retaining staff in AI machine learning when you're paying government salaries. It's tricky. To put this in perspective, we just lost a potential candidate to Zillow because they said they'd pay off his student debts and

start him on 200,000 a year. Tricky for government, to say the least.

Moving along. Biocompatibility/Tox is always an area that we have a lot of interaction with NCTR, clearly. And we very much value our collaborations with NCTR. It's an ongoing area, without a doubt. Our fundamental aim in biocompatibility is to try and be able to do that from a theoretical standpoint rather than having to do actual testing to do it. And that's an area we're moving towards. It involves computer modeling as you'd expect. But it also fundamentally involves an understanding of materials. And I'll come back to that in a little bit. Biocompatibility for us at CDRH is probably, I think if you took a straw poll, would be the biggest headache for most people. Of all of our 25,000 or so premarket packages that we receive each year biocompatibility raises the most deficiencies in those premarket packages compared with anything else at all. So, it's clearly an area that we've got to get a handle on. We're making a lot of progress and I think our collaboration with our colleagues down here really helps that area as well, of course.

Physiological closed loop systems are a thing again. There are some of these that are evolving rapidly and that again is a theme for devices. Closely monitoring

systems traditionally have been things like heart/lung machines. They're now getting much, much, much more sophisticated. Obviously, AI machine learning has a big part to play in these as well. But machines like this are actually now making diagnostic decisions and treating the patients. So, we have a lot of work ongoing with our colleagues at CDER in this area, because if a particular device, capital equipment device is making a decision about treatment, is that treatment on-label? Is it off-label? And that again, introduces really exciting questions for us.

Computational modeling is something that - as a molecular biologist, you can imagine how much I know about computational modeling - but it is an area that I think that the FDA is leading the world, certainly the world of regulatory science. We have a fantastic Agency-wide Work Group that does all of these areas of computational modeling enormously successful. Every single Center is represented.

As an example of the output of that, about 12 months ago we published the very first fully in silico clinical trial, in the Journal of the American Medical Association, that compared two different types of breast cancer imaging. The real trial - because, of course, we

had the data at CDRH - involved I think about 300 or 400 patients who are getting double exposed to CT and digital breast tumors synthesis. Six years to do the trial. We did it on 10,000 patients in a weekend and got the same regulatory result. So that's been published in JAMA about a year ago. And the dataset we generated from that and the algorithms are public domain and they're actually getting downloaded at the rate of about 700 a month at the moment. So, we're really proud of that because we're giving back to the whole community on that one.

Digital Pathology, again, is an area for the future. We have cleared two digital pathology devices at this point. This is an area where we are doing an awful lot of work in terms of trying to generate huge annotated datasets for different cancers and things like that. This is an area that I anticipate we will be having a lot more interaction with the National Cancer Institute at NIH. It's a hugely important area. We're actually trying to stimulate that innovation in a huge way, where we can.

Electrical Safety is something that is important for us. Probably not the other Centers quite so much. Again, it's one of those things that's crossed over from the consumer area and certainly affects medical devices.

If a battery goes pop like it does on some laptops and things like that.

The big couple of areas I really wanted to spend a little more time on, Materials Performance. As a Center, we regulate the entire device. The question we ask is, is the device safe? Is it effective? And things like that. However, every device is clearly made up of specific materials. And this is an area that is very much in the public eye at the moment. And we have an awful lot of interest from the outside world and we welcome that very much. It's a debate that's not going to go away very quickly, because essentially it transpires that even though many, many devices have been implanted for many years, and they have a fantastic benefit for those patients, we don't actually know as much as we would like about the performance of those materials in the body.

Metal alloys are a great example of that. We recently had a public workshop that looked at metal alloys and metals in general as well as dental amalgam. And that was a very, very vigorous public debate. We welcome that. And we're very much in listening mode on that whole area right now.

Couple of other areas I want to mention before I think about the future a little more. Nanotechnology, I

think is an area I would highlight as being a fantastic collaborative area between Bill's group down here in Arkansas and ourselves up at White Oak. Within CDRH, we house what we used to call the Nano-Core, now called the Advanced Characterization Facility, very similar to the group that NCTR has down here. They act as one team together and it's a pleasure to have them. They really are a genuine resource for the whole agency, I believe, and they do a wonderful job.

The last one I want to mention before I move on is therapeutic ultrasound, because it embodies some of the things I want to think about for the future, which I'll get to in a second. Therapeutic ultrasound is currently used very, very high-intensity ultrasound, and it can be used for tissue ablation, it can be used for noninvasive removal of cancers and things like that. We recently, over the course of this summer, qualified our first medical device development tool in this area, which is a tissue mimicking phantom. It allows you to actually assess the thermal properties of your high-intensity therapeutic ultrasound device in a way that we qualify as a tool very similar to CDER's Drug Development Tool Program. This was our first one on a non-clinical assessment methodology.

This actually sets the cornerstone really for how we want to move forwards.

Industry now has a common tool that they can all use to assess the thermal properties of tissue if they're going to develop a high-intensity therapeutic ultrasound device. As of right now, there are six different SBIR companies just through the National Cancer Institute developing new high-intensity therapeutic ultrasound devices. So, by us being able to provide a standardized tool for assessment of those devices, essentially removing things from having a ferry from one side of the pond to another, to building a highway straight across. And that's fundamentally what we are trying to do at CDRH through our Reg Science Program, is to increase the efficiency of future regulatory processes.

So, I've talked a little bit about the materials performance side. I'm not going to go on about that, particularly at the moment. It's really a challenge that we have within NCTR and ourselves for trying to understand the at-risk populations. And I think the more we're learning about specific materials, the more we know that there are certain subpopulations that are more inherently at risk from certain types of material if they're implanted for a long period of time. This is the beginning

of a new journey to try and understand a lot of these materials. And I think ourselves and NCTR will be absolutely at the forefront of that understanding.

In devices, the vast majority of innovation comes from very small companies. Ninety percent of people in devices actually work for companies of 50 people or less. And what we're seeing is a continuation towards larger companies actually having a shopping list rather than driving a lot of that innovation themselves. There are some notable exceptions, but a lot of the time that innovation is coming from small companies.

So, what we're trying to do through CDRH is to do what we can to keep this company alive. Standardized tools are clearly one way of doing that. But what we are actually trying to do in the future is to partner much more with the NIH to say, well, let's work on it together. You're putting a lot of money into fundamental research, which is a great thing. Putting a lot of money into SBIR companies, which is a wonderful thing. But what happens at the handoff between each of those phases? That's where a lot of that innovation is at risk. That's where it falls down. So, we're going to actively work a lot closer with NIH as we move forwards to actually sort of not paper over the cracks, but to help people lead through those areas,

because that's where technology fails. And we have a fundamental, not just objective, but a responsibility within CDRH to get those companies to survive longer, because that's where the new technology is coming through to the market and to the patients from.

So, I'm actually going to finish with just one summary slide. Part of our mission at CDRH is to stimulate innovation. One area that we can do that is, as I said, by pushing forward with translational medicine, but it's very difficult to do that in the current climate. So, we're actually trying to turn ourselves within my office to be an area that qualifies tools for future use. If we can then have a standardized set of tools, that gives the small companies a much, much better chance of survival and that stimulates innovation.

We're also starting - many of you may have seen specific innovation challenges. So, we've had one recently on sterilization of medical devices. There is quite a lot going on in the outside world concerning ethylene oxide sterilization, which I don't want to get into right now. But there have been one or two plants that have shut. What that means is medical device shortages. And medical devices save lives, like all the things that we work on at FDA. That's really one thing that binds us together. So

we're trying to stimulate a lot of those innovation pathways by saying, if you have good ideas in this particular space, then we'll give you a certain amount of tender loving care when you come inside and we'll work with you. And we're trying to do that a great deal.

And as I've said, the other angle that we're really trying to address is to understand better what happens to materials as they're in the body for a long period of time. So, I shall stop there, if that's, Greg, all right, timing-wise, and invite any questions if there are any.

DR. LANZA: Thank you very much. It was great. If I could take the Chairman's prerogative and ask the first question. We have about two minutes. I'm Greg Lanza. The question I have is, have you considered, given the 5G situation, the transfer of medical imaging data, especially when it involves a combination of imaging data and demographic and medical data as AI starts to integrate these prospectively for toxicology, for instance? Transferring that to a second machine at a different institute, it could be the same vendor, possibly another vendor like where you transfer DICOM, because it allows patients to not have to go a long way for care but be part of a serial program. And the implications for that besides

the data transfer implication on regulatory, I wondered if you've thought about this.

DR. MARGERRISON: We're beginning to right now. It's a really, it's a fascinating area. There are different aspects that we're involved with as a Center. There are things like interoperability, because if your data does go to a different site, the format of that data, et cetera, et cetera, is so important. Data integrity, personal PPI protection as it's going, that's all super important. But my eyes were opened actually not that long ago by one of my staff who showed me a video which is on YouTube. And this is a live demonstration by Wahwa(phonetic) in China who did real time surgery. The surgeon was 30 kilometers away from the surgical robot and he was operating in real time with under 100 millisecond delay through 5G.

So, what that means to me is not just you can take these big data chunks everywhere, but as of right now, everywhere in a medical device where we've got information transfer can now happen at a remote site. That has enormous implications on us. That's one of the big areas. When I talked about wireless compatibility in my program areas, yes, Wi-Fi interference in the hospital is important and we continue to look at that, but actually,

implications for 5G are much bigger than just a quick network. And another area that we know, some of the questions we need to answer, but not all of them right now. And I think that's the essence of regulatory science, is trying to have the best crystal ball in the world.

DR. LANZA: Any other questions? One last question. Ken.

DR. RAMOS: That was fascinating. To what extent are you looking into cybersecurity issues and interfacing with cybersecurity in the context of devices, particularly?

DR. MARGERRISON: Enormously. It is an area that is - and there is actually one public example, which, yes, it is public, where a group of people hacked into a pacemaker to try and prove that it was possible. So, the share price of the company went down and they could short the stock. That genuinely happened. It is a hugely massive, massive area for us. We have guidance that we've put out there on it. But fundamentally, it is a very difficult thing for us to check in a premarket application, actually, because we can't have a team of white hat hackers sitting there trying to get into these things. But there are very established guidances and procedures that we expect manufacturers to follow at this

point in time. What we're finding is that the manufacturers are very happy. The vast majority are working with us, without a doubt.

The issue we've got at the moment is more which devices that were already out there are ones that are susceptible. In some ways we're more worried about those. In the example I just mentioned of the pacemaker, even though the manufacturer developed a software patch really, really quickly, there were some patients it couldn't be applied to because you can't just stop a pacemaker with all patients for obvious reasons. So that was quite an interesting example.

But we have a whole dedicated division just to cyber security. Oh, yes. And we work very, very close with all the leaders in the field. And the vast majority of Medtronic being a great example of a company who have a lot of devices that potentially could go wrong. The other angle that sometimes is forgotten about cyber is we think about the baddies out there trying to hack into things. Sometimes it's the patient's own families who do that. There is a group of people called loopers who take their diabetic insulin pumps and they think they can download a better piece of software from the internet to run it than has been developed by the company that produces it. So,

they hack into the phone or the device and actually upgrade software to something they've got off the internet. So that again, is a cyber security vulnerability, but meant for very different purposes. So, again, very different angles to a lot of these things.

(Off mic comment)

DR. MARGERRISON: Absolutely. Enormously. It really does. Yes, and we have a whole dedicated unit to exactly that. It's the big headache of our times.

DR. LANZA: And with that, we're right on time. Thank you very much. And we'll go for lunch.

(Lunch Break)

Agenda Item: Center for Food Safety and Applied Nutrition

DR. LANZA: - session scheduled from 1:00 to 2:00 for public session. And that's going to not occur because the speaker didn't come. So, the we're going to begin then on the schedule at 2:00. And this is continuation of the discussion from the presentations from the different Centers. And the first presentation is going to be the Center for Food and Safety Applied Nutrition. And this is Suzy Fitzpatrick.

DR. FITZPATRICK: At the Center for Food Safety and Applied Nutrition we have a lot of different compounds. We go from cosmetics, contaminants, constituents, indirect food additives, direct food additives, color additives, and cosmetics. But the one thing about our compounds are we don't have preapproval authority over anything except color additives, indirect and direct food additives, which is indirects or packaging material, and color additives, direct food, and color additives. Other than that, everything we regulate, which is cosmetics, dietary supplements, contaminants, constituents, which are byproducts of heating that you find in food, we have only post-market approval. So, what we have to do in order to do anything about any of those

compounds is we have to show some type of harm, either how they're being used ordinarily or by their label.

What that means is, since the sponsors of those compounds aren't willing to do the research for us, we have to do the research. And so, we're very dependent on what we do at NCTR as a very strong partner with us in working with - and in fact, all of our animal research is being done at NCTR right now, along with some of our alternatives.

So, RADM Hinton mentioned the Predictive Toxicology Roadmap. We assist and chair this Committee because it's very important to us because as I said, as we're generating data, alternatives are important, since we sometimes need data very quickly. And in areas where there's not a lot of animal work or we don't have time for the animal work.

So, what we have done is we've looked at - after we worked on the Roadmap, some of our tox studies that - what we are using now, we look at studies, we looked at the bioassay, chronic bioassays and we looked at the use of the dog for chronic testing. And then we're evaluating some new tests that can help us.

The first thing we did was we looked at the bioassay, toxicity study in dogs. It's just traditional

that you do a rodent non-rodent species. It wasn't really ever based on any type of scientific discussion. In fact, if you go back to what was called the Black Book, which I think came out in 1949, which was tox testing in foods, they had to two species, but there was no real scientific reason for it. It was, well, we might miss it with one, so we'll get it with the other, rationale, sort of more of a reliance on that.

And so, we decided, do we really need the dog study for food and color additives? And what we did was we went back and looked at 160 - all of the studies we had on the dog to see if there were any unique endpoints for food and color additives or if we could have just used the rat and approved the compounds. We found out of the 164 studies there maybe were five where we set an ADI on, but then looking at what the rat, what the ADI was for rat, we would have approved it with the rat data. So, we've decided that for food and color additives we do not need a dog study. We don't have a requirement, but we don't need to get a dog study. We can look at a "second species might be modeling". And this is where we probably come to NCTR to help us look at what type of modeling can actually give us better information on the fate of different chemicals in food. So, that was one thing we worked on.

And the other thing we worked on was starting to talk about the rodent bioassay. We had an FDA and SOT colloquium last February. We're also going to be working - we have one with Eurotox at the 2020 Eurotox meeting in Copenhagen. We are presenting something at the Winter Tox Forum. This is something we were working on together with EPA, which is also presenting, and with an NIEHS and (indiscernible word). What happened with CFSAN was we just had to take seven synthetic flavors off the market because high-dose testing showed that they were carcinogen in high doses, even though at doses that you would be exposed to, they would not be harmful. But because of the law, the Delaney Clause, we had to remove them.

So, for instance, one of them was ethyl acrylate, which the lowest dose was a million times higher than anything you'd ever be exposed to in food. So, the first seven we took off were kind of inconsequential, but it's going to apply to a lot of your favorite flavors pretty soon if this trend continues that they're using the high-dose testing to inappropriately remove actually safe compounds from the market. And it's very difficult as a scientist to not be able to use the best possible science to regulate compounds and to go with antiquated laws. So, one of the things we would like to see is a change in how

bioassays are run, that maybe instead of looking at hazard, first you look at exposure. And then like CDER is looking at, you decide the doses based on exposure, calculate exposure to a compound instead of using the MTD or some other traditional way of doing so.

This is one thing we're really interested in. We're working with ICCVAM also because they want to kind of almost immediately go to some of the high throughput testing and we're telling them that all of these tests, any animal tests or any alternative are really tools for a regulator to answer regulatory questions. And you can't get new tools unless they answer the questions that you need answered to regulate the product. And so, we volunteered to take some of our cancer people that work on bioassays to ICCVAM and say, as you're looking at alternatives, these are the questions you need to answer. These new studies have to answer these questions because our job is really to put safe and effective products on the market.

So, the next thing we're looking at is read-across, which is an alternative. We're looking at read-across. And then this expanded decision tree approach, which is the TTC, Threshold of Toxicological Concern. So,

it's confusing cause the little box is the one that's up there and the big box is here.

So, we're going to put together a cooperative agreement probably with Underwriter's lab at FDA and CFSAN to look at their read-across tool. This is where we're going to come to NCTR because we're going to be testing it to see if it works for DART. The endpoints in their tool now are these endpoints. But the DART one is under development and will be interesting to see with some of our colleagues if this new tool can give us some DART information. This wouldn't be for regulating a compound but would help us screen different compounds because like we said, we have a lot of compounds. And you know how many supplements are on the market in cosmetics and contaminates on the market that we need screens to kind of parse of the ones that we want to look at or not.

The other thing we've been working on is expanded decision tree approach. Originally, we had FEMA, which is not the place that helps you when you're in distress, but it's the Flavor Extract Manufacturing Association. And they put together this because flavors could - some of them are used so little that you really can put them without any data. So, this is a TTC approach to characterize them first into what there were three

categories, but we have expanded the decision tree approach to put things into six different categories to categorize this based on the level of concentration that you can see in the product and what type of data that you need on it. And if you look at how we do packaging material anyway, it's a tiered approach where depending on the concentration, really the estimated concentration of that packaging material that can migrate into your food, that's how we decide how much data we need.

So, this is an area that both Donna and I have been talking to the EU Tox Risk Group that is now coming up to redo their EU Tox Risk program. They got a five-year grant to look at alternatives under EU Horizon 2020. FDA is joining them for the next session. And they're interested in this TTC approach, and expanded decision tree. We use it for excipients. I think CDER uses some TTC approaches for contaminants, I think, or in pharmaceuticals. So it's really a good way to actually eliminate a lot of animal testing, because if you fall into one of the categories that are so low that you're not concerned about it, it's another - and this is the expanded tree. You can see it's got a lot of different questions. It's got 160 questions. So, it's probably

rigorous to use, but helpful or maybe take less time than doing an animal study.

The next thing that we've talked about that we're working with, we have a strategy for toxic elements, which is what we call metals. And metals are considered contaminants. They're in food. And we've been criticized a lot about - especially recently Consumer Reports came out with the levels of different metals in infant and children's food. And knowing all the metals of developmental neurotoxins, this has been something that we've been focusing on.

The key features of this is to - this was Dr. Mayne, which is the Center Director's Program - is to prioritize then make decisions on it, and then look at science and then at the policy. So, it's kind of an elaborate thing, but it's a way of deciding which metals we're really focusing on. We were part of the Federal Lead Strategy Group that all of the Federal Agencies participated in for the last few years, chaired by HHS and EPA. And actually, they're having a meeting today and tomorrow on their Lead Strategy and CFSAN was part of that looking at changes - allowable levels of lead in children's food. So, we lowered by half the allowable levels of lead in children's food and also lowered the

amount for women of childbearing age. So those aren't safe levels because there are no safe levels for any metals, but they are levels that are one way of containing the exposure.

The other thing we were interested in is mixtures of metals in food because what we look at right now is one commodity and one contaminate at a time. But we know that's not the way you're exposed to mixtures of metals and contaminants in your food. So, one of the things we were interested in was. For all the neurodevelopmental metals that are in baby food, how do they interact with each other? So, we looked at the C. elegans and we saw that some of the pathways that we knew arsenic affected were conserved down to the C. elegans model and we used that to design a developmental activity test.

And at the same time at NCTR, we have a study going on with Sherry Ferguson to look at the effects of development exposing in utero and perhaps to inorganic arsenic and how that affects cognitive behavior. And we found that some of the results that we found in the worms, in other words, developmental delays and effect on cognitive behavior, motor function, are the same things that Sherry is seeing in her inorganic arsenic studies,

which was really exciting because it's the same things you see in the Epi studies from populations that are exposed to large levels of inorganic arsenic in drinking water. And so now, Sherry and I were now just talking about expanding this to look at zebrafish and see how much zebrafish can give us on metals alone and mixtures of metals for developmental, neuro, and for cognitive behavior. So that's an exciting study. And we might be working on it with the Environmental Defense Fund, surprisingly enough, who has gotten some money to study this at Cornell and wants to work with us on the zebrafish.

This is my favorite technology, organ-on-a-chip, because I was there when we first started on it. It's like my baby. And I was there when DARPA came into the Office of the Commissioner and said, I'd like to develop organs-on-a-chip. And they had a picture of a box and a funnel and an arrow coming out. And so, FDA worked - all our Centers, worked with DARPA and with NCAS to develop the first five-year program. And then FDA CFSAN had the first CRADA here at FDA to bring the organs-on-a-chip technology into our laboratory. So, we developed a CRADA for the liver on a chip. They brought it in here and now we're expanding that. So, Donna and I worked on a larger CRADA

where we're bringing in organs-on-a-chip to go to the rest of the Centers from Emulate. So, we're talking about ones for CDRH, for maybe biologics for that. So, we're about finished with the CRADA. That's going to be signed not by the Office of the Deputy Commissioner for Foods, but by Denise Hinton. We like to keep her busy. And then we'll be available to talk. And I'll talk a little bit later about how this fits in our overall program on alternatives.

The last thing I'll talk about is a lot of people have talked about - and of course, with Donna being in charge of this, the research of organ-on-a-chip, that's very much centered through NCTR and our collaborators. And again, Donna and I represent FDA on the Tox 21 Partnership, which is a partnership between us, EPA, NIEHS, and NCAS. We've often been asked, how does high throughput screening even help FDA? So, we haven't really looked at it till now. But right now, my ORISE person is looking at with EPA and with the Office of Food Additives, is looking at how the PODs, that EPA is calculating from their high throughput screening, match the data we have on those compounds in animal studies.

So right now, they're finding that it's not always consistently - they're always much lower but it's not really - we haven't found a consistent pattern yet,

but we're persisting because a lot of this is especially useful for cosmetics, because, as you know, cosmetics in Europe aren't supposed to be tested in animals. There are two or three bills now in Congress to extend that to all cosmetics. And we do have some animal cosmetics studies done at NCTR, so we'll have to hurry those up because we will all be not permitted to do any animal testing except if you get a waiver from the Secretary.

So one of the things we want to find out, is the high throughput screening at least able to give us some prioritization for dietary supplements or for our cosmetics so that we know where we should do testing or not and can be a little more focused. This is an exciting project for us because we're at least considering how this new technology will work for us.

So, Denise mentioned briefly we started an In Vitro Systems Working Group that I chair and co-chaired by Donna to look together as an Agency moving toxicology more towards more predictive alternatives. And so, we want to make sure that our regulatory scientists are up to speed on alternatives before they seam in an application. Our research, including all of our research at NCTR, will help us focus on alternatives and leverage what we have there and then to develop potential public/private partnerships

that we can use to develop going forward. And we are working on a couple of public private/partnerships. I know I have almost completed one with FDA and the University of Illinois and with the food industry to look at predictive toxicology for the food industry. So that's Bob Brackett who used to be our Center Director, is doing that. He's almost got all the SOPs together, along with a major part of the food industry to move that one forward. And then we have a couple other ones that we're working on.

Our first test case will be organs-on-a-chip. All of our Centers have organs-on-a-chip. A lot of us research Centers, CDRH is developing one. CBER has some very nice organoids that they're working on. CFSAN has the Emulate Chip. CDER has the liver on a chip from CN Bio, which is an MIT spinoff. NCTR has the tissues chip, which is the German chip that's used almost exclusively in Europe. And we're figuring by getting all of these people together, it will give us enough exposure so that we can move forward with development performance standards for organs-on-a-chip.

FDA doesn't endorse proprietary technology, but we do develop performance standards that can be used to evaluate chips. And so actually, CBER just sent us today a draft definition - our first thing was to develop a draft

definition for what organs-on-a-chip and microphysiological systems are that we can then share with our stakeholders. And CBER was working on that and just sent that forward to all of us to work on. So, it really is a collaboration between all of us. And having the partnership, the expanded CRADA with Emulate moves into that. We're also talking - Donna and I are talking with Tissues and with MIMETAS, are two other chip companies that we might get some chips from there, and trying to really give us a lot of experience so that we can know about this technology, because we're really being asked from the outside - really having the outside trying to tell us, this is what you should be doing in this area. And we feel that we probably know best what the requirements are to move this type of technology into regulatory science.

Just to mention some international activities, the ILMERAC, which is International Liaison Group for Methods of Risk Assessment for Chemicals in Food, is an EFSA collaboration globally that we're part of. And EU Tox Risk, as I said previously, they want to engage more with FDA in their next rendition of this, especially on organs-on-a-chip and TTC. But they were also looking at repeat those toxicology and DART for that too. And that's

something Donna and I are moving NCTR toward, but probably be very, very heavily involved in this too, as our premier lab.

And I forgot to mention a couple of other things because I thought that they would be mentioned previously. We do have several studies at NCTR and several more exciting studies to come. They are doing all of our cosmetic research and I think Gonçalo will talk about the research on tattoo ink. So, tattoo inks that people use were not developed for human. They were developed for your car and that kind of stuff - industrial use. But they are then used in people. And we've measured a lot of those tattoo inks and found they're full of contaminants. So, it's not just the ink or the pigment, it's PCBs, it's lead, is that kind of stuff. And they are considered cosmetic. So, i.e., they're not regulated because cosmetics aren't regulated in any country.

Previous work at NCTR demonstrated that a lot of those inks, maybe 40 percent, I think, become systemic. So, they come systemic and a lot of them go into the lymph nodes. And whether they're in the lymph nodes doing anything or they're just getting in the way, we don't know. But we also assumed that they're probably crossing the placenta and going into the developing fetus. A lot of

women of childbearing age do have quite a bit of ink on them. So that's one study that we're having done under Bill's DART program to look at where those tattoo pigments go. Do they cross the placenta? But also, how many are going through the blood brain barrier into people, either to the mother or the child? So that's an exciting study that we're looking at.

We're hoping to also look at nano and micro plastics there. We know that they've been measured in some food commodities, especially in fish. But we have yet no idea how small the particle has to be to cross the stomach and it go systemic into the person. And then, we assume nano can cross into the cell. But again, what is it doing there? So, we're looking to the Nano-Core at NCTR to really help us on that. That's a really important and really, really big developing area that is going on that's just going to get bigger. And already O'Neil(phonetic) has set up an Agency Working Group and linked us into the Global Working Book. They had the Regulatory Council on that. And so that's something that hopefully we'll have more information to tell you next year about it.

The other thing I didn't mention, the biggest thing right now besides BPA, which we won't talk about, is perfluorinated compounds. And NCTR is looking at the

shorter chain, the C6 perfluorinated compounds, which their packaging people are moving to. They're doing some DART work on that to find out if they're as hazardous as the longer chain ones. So, I think just to say, we are really pleased at CFSAN with all of our collaborations with the NCTR. They work with us on the protocol. They keep track of it as they go along. They really are truly been partners in research. And I just can't have enough gratitude to all the things that they've given us to help us regulate the products that we have. And we look forward to having more partnerships in the future.

To just say we are moving towards alternatives. We look at NCTR to help us make that very important move in toxicology and make sure that we compare them with concordance stated in animals. I'll take any questions.

DR. LANZA: Thank you very much. Since we cut an hour, we have a couple minutes. I'd like to keep it to two or three minutes for questions. If I could ask one, what perfluorocarbons are you talking about? The perfluorobutanes, the perfluoroprotanes, short chain versus say the (indiscernible -- cross talk) boiling points or -

DR. FITZPATRICK: The C6 is what we're looking at. The C6 perfluorinated compounds that are in packaging.

They're in anything like a pizza box or popcorn, inside the popcorn. Anything that's kind of greasy in food, you might have some of those because they're degreasers.

DR. LANZA: Thank you. Thank you very much. The next speaker from the Center for Tobacco Products is Dr. van Bemmel.

Agenda Item: Center for Tobacco Products

DR. VAN BEMMEL: Good afternoon, everyone. I'm Dana van Bemmel. I'm the Branch Chief of the Office of Sciences Research Branch. We're within the Center for Tobacco Products. I think I shared with you last year that we are not on the White Oak campus. We've moved off campus to the Calverton Building. But the Office of Science is the only office that's part of the Center that is offsite. The rest of the Center for Tobacco Products is at White Oak.

I decided to take a little bit different slant this year on our update. I'm not going to talk quite as much about our current research portfolio, but many of you know me and you know that I'm very proud of our research portfolio. I'm happy to talk to you at any time about any of the other types of research that we're funding within the Center for Tobacco Products. But today, I'm really going to focus on an update from CTP. Just a brief

reminder of who we are, what we've been doing, some of our regulatory activities in the last year and then some of the research that we have completed with NCTR and projects ongoing and where we're looking in the future.

So, I've heard several Centers actually say today that their Center is different than the other Centers at FDA. And I usually say that too. So, I guess we all in some way feel like we're a unique Center. I will say that we are only 10 years old. We were established in June of 2009. And just as a reminder, in that first iteration, we had the regulatory authorities, FDA was given the regulatory authorities to regulate the manufacturing, marketing, and distribution of tobacco products. That included cigarettes, cigarette tobacco, roll your own, and smokeless.

And then in 2016, FDA finalized what's known as the Deeming rule. And the Deeming rule then brought into the regulatory realm of FDA tobacco products excluding their accessories such as electronic devices, so e-cigarettes. It also deemed or brought into Center for Tobacco Products, regulatory authority, cigars, pipe tobacco, nicotine gels, water pipe, dissolvables, and any future tobacco products. And so now all of these fall

within the Center for Tobacco Products regulatory authorities.

The Center for Tobacco Products has a number of different regulatory activities that we perform. We have premarket review of new and modified risk tobacco products. We have post-market surveillance. We can implement product standards. I talked to you about some of that last year and some of our advanced notice of proposed rulemaking around nicotine, for example, reporting of ingredients, reporting of adverse events, health warnings, advertising, promotions, and all of this is based off of user fees.

In general, CTP does not regulate a number of different things, and it's outside of our regulatory authorities, and I like to always mention this because once we get into a discussion of the kinds of research or the kinds of activities that CTP should be thinking about or doing, often things such as clean indoor air policies or changing the minimum age to purchase tobacco products comes up. And so, as you can see, there are a number of different things that just simply fall outside of the Tobacco Control Act or outside of the law, so are outside of our purview.

CTP regulates tobacco based on a population health model, and I do think that this makes us different than other Centers with that at the FDA, we know that tobacco is inherently unsafe, so we can't use the traditional safe and effective standard when reviewing these tobacco products. So, it's really a regulation based on looking at the risks and the benefits to the population as a whole. And you're going to hear this mentioned throughout my talk and probably throughout any talk that you might hear from someone from CTP. But it really does make us uniquely different. We're looking at the population as a whole, and that includes both users and non-users of tobacco products. And how what we might do would impact that group.

I am the Branch Chief of the of the Research Branch, and so I spend most of my day thinking about regulatory science, but what I found when I first took this position in 2011, I wasn't the Branch Chief at that time, but when I joined CTP in 2011, was that a lot of folks didn't understand what regulatory science was and they didn't understand how or why that was different than the types of research that was funded at NIH, which is where I had come from at the National Cancer Institute. And so, we spent a lot of time in the early years of the

Center talking about what regulatory science is and how it itself as a discipline is different. It's not just about innovation. It's not just about understanding the mechanism, how the damage actually happens, how the protein bonds might break, but it's about looking at research that's going to inform our regulatory activities. And I'm going to touch on some of what those are, at least for our Center specifically.

But it's science, any science, any research, any data that's going to inform our regulatory activities. But sometimes it's not the most exciting, the most novel, the most innovative research, but it's research that needs to be done in order for us to evaluate those products. And I think you'll see some of these references actually are from previous commissioners. And I think that idea of regulatory science is - that that part of it is not unique to CTP.

So, as I've mentioned, the Center uses science in all of its regulatory decision-making processes. And so instead of going through some of our research portfolio and highlighting the number of projects, et cetera, I thought I would talk a little bit about some of the regulatory activities that have been going on over the last year or so.

You may have read in the news that CTP recently authorized the marketing of a new tobacco product. This is the IQOS Tobacco Heating System. It's an electronic device that heats tobacco filled sticks wrapped in paper to generate nicotine containing aerosols. It initially hit the market this past August in Atlanta but has been authorized for marketing across the United States.

And in authorizing these products, what FDA is saying is that these products were found to be appropriate for the protection of public health, because among several key considerations, the product produced fewer or lower levels of some toxins than combustible cigarettes. I will say that the authorization is rather lengthy. It's really interesting if you have any interest at all in tobacco regulation or in the IQOS device, I would encourage you to look at the authorization. I also encourage our researchers in the research community to look at the authorizations because anything CTP puts out is a hint or is a way of communicating the kinds of research that we are interested in and continue to be interested in funding.

So, in this case, there are stringent marketing restrictions on the products to prevent youth access, use, and exposure and post-marketing requirements for

monitoring market dynamics. Again, such as potential use uptick. And so, these are the kinds of surveillance and studies that will be important to the authorization of this particular product, moving forward.

FDA is also working on a number of different rules and foundational guidances. I don't necessarily want to read each one to you specifically, but I would like to highlight that these all indicate various pathways for tobacco products to come into CTP to be reviewed and potentially enter the marketplace. So substantial equivalence, I think I've spoken with this group about before, but that's a pathway in which a product could come in and be reviewed as to whether or not it's substantially equivalent to a predicate product on the market. The Premarket Tobacco Product Applications, or PMTAs, as it will be noted in the rest of the presentation, for a number of different products, including modified risk tobacco products as a third pathway. This is a pathway instead of being premarket or new, it would be a product having a modified risk claim and then of course, tobacco product manufacturing practices.

We have been trying to and will continue to engage all of our stakeholders, so researchers and industry alike, to work through our processes because

again, we're relatively new. So unlike Centers that have decades of guidances and policies to lean on and fall back on, we are developing many of those, as you can see here. And one of those ways that we've been trying to do that include public meetings. So, this past August, we had a public meeting to talk about deemed tobacco products and their pre-market application process.

I also just wanted to note, I don't want to go through all of the details of this, but this past summer there was a court order for all deemed tobacco products that were on the market as of August 2016 that they must file a pre-market application within 10 months of that order, which is essentially May of 2020. And so, what this means, and why I share this with you, is that one of the major activities that is and will continue to happen within the Center for Tobacco Products is pre-market review or PMTA. We anticipate that in the spring we will have a large number of applications to be reviewing. So, any kind of research that we have, tox research and otherwise related to these deemed products is going to be very important to the activities happening within the Center.

I said it before, and I'll say it again. Data drives everything. Data drives the decisions that help CTP

achieve its mission of reducing morbidity and mortality associated with tobacco use. My kids get tired of hearing me say it, but science and data is everywhere and it's exciting. And in this case, we truly depend on it.

We have a number of research priority areas that we have published on the web and in other places. Currently we have eight research priority areas. I did not list all eight here. I just pulled out toxicity because we are here at NCTR. But I'd encourage you to look at those online if you're interested. But when we talk about toxicity as a research priority area, we're really talking about understanding how tobacco products and changes to the tobacco product characteristics affect their potential to cause morbidity and mortality. And those include both animal and subculture models as well as novel alternative tox model approaches that test the toxicity of tobacco smoke, aerosols, or specific constituents in the tobacco.

I've highlighted some of our specific areas of interest. We do update our research priority areas. We try to at least update them every year. It falls more about every year and a half, two years. But we revisit these annually to make sure that the research that we're funding and we're continuing to engage in is supporting the mission in the high priority areas of the Center.

In the next few slides, I'd like to just talk to you a little bit about some of the research projects here at NCTR that we've completed. This is by no means an exhaustive list, but it's just a sampling of the types of research projects that we have funded here with NCTR.

The first was completed in 2014, priority setting of harmful and potentially harmful constituents in tobacco smoke products with bioinformatics. And so, this was really a project that helped us support priority areas around tobacco smoke constituents and decision making on whether or not those constituents required for their evaluation and/or could inform a product standard. And what they found in this particular project was that 47 percent of tobacco smoke constituents had limited scientific data. So, there is a lot of other tobacco research has been around for many decades. There's still research to be done even in tobacco smoke.

Another completed project here was the extrapolation of in vitro acrolein dose response derived in air-liquid-interface airway epithelium models to in vivo lung toxicity. And you'll see this as we get towards the projects that are current, but we have really three major areas of research here with NCTR. And I think Bill touched on these earlier. One is around the inhalation

work that we do in animal models. One is around the ALI or air-liquid-interface work that we're doing. And then we have bioinformatics and modeling. And so, this one that was just completed in FY18 speaks to some of that modeling work.

We did relatively recently complete a 14-day nose-only inhalation tox study of NNK in rats. It was finished in an FY15, I guess. This particular study gave us information around the dose range of NNK in the subsequent studies where the tox of NNK following this for the following 90-day repeat dose study, which was designed and later conducted. So, you can see that we have some research that I would really consider more methods development and informing the next steps of our research program and portfolio here with NCTR and other collaborators.

I can't remember if I mentioned this. I apologize if I did. CTP does not have labs of our own, so that also makes us unique from other Centers. So, all of our research that we do is in collaboration with other partners. So, we do a lot of our toxicological research here with NCTR. We have a large portfolio with NIH where we're funding grants. We also fund a lot of research with other federal partners such as CDC.

Some of the active research projects that I wanted to highlight here include the evaluation of toxicity and inflammation produced by cigarette smoke. And this is using the in vivo airway models. This is set to be completed in fiscal year 20. The benefit to FDA and CTP with this particular model is that the hope that the data generated from this will help inform ALI model will generate data that will have better predictability of in vivo responses than other in vitro or rodent-based systems, and help us to inform what we are planning and other regulatory activities and to help us understand the potential risk related to those activities and new products.

Aerosol inhalation exposure chamber development is a current project as well. This is obviously with the inhalation group. This is really a project looking at developing a tiered inhalation exposure system to deliver equivalent aerosol rates across all exposure levels. These simulations are hopefully the data generated could inform optimization of the inhalation exposure chamber, leading to additional dose response and specific research questions around specific constituents or other tobacco product exposures. So, this is really building on the work that we've already done with the inhalation group.

Just a few other current projects include some PK studies. Some of these are building off of projects that were recently completed using some of that data to fill in some of our other research gaps. And when I think about our research portfolio, where we are now and where we want to be in the next one, three, five years, there were three real areas. I think, again, in those three bucket areas that that I talked about earlier where I could see us engaging with NCTR on additional research.

The first, of course, would include the inhalation tox studies, the evaluation of toxicity with repeated nicotine inhalation exposure from tobacco would help inform some of our regulatory activities. And in addition, determining the inhaled nicotine concentrations with acute nicotine inhalation exposure that may or may not lead to significant adverse health outcomes could help inform future regulatory activities at CTP.

The ALI Group is I think is another group that we would hope to continue to work with. Some potential areas of further collaboration I could see would include determining cytotoxic and genotoxic potential areas, areas around aerosols generated from electronic cigarettes or ends. Looking at genotox and cytotox potentials around the aerosolized ingredients that might be unique to different

end products. And then simulation of human inhalation exposure with ALI cultures and epithelial tissue models. Again, I think all of these would fall within that ALI core group.

And then finally, CTP is interested in flavors in tobacco products and the chemicals involved in flavoring a tobacco product. Some additional areas of research I could see here but include toxicity that may result from chemicals formed when a product is heated or burned. And studies that would inform not just that, but what toxic chemicals might result from the heating or burning from these flavors are chemicals and it might be a specific chemical, or a group of chemicals related to that flavoring. In addition, on the potential toxicity or adverse health effects from exposure to these various chemicals when they may be heated or burned or aerosolized.

So those are just three sort of general areas where I could see us continuing some of our collaborations. I will just say, like many of my colleagues from other Centers at FDA, we've had a great working experience with NCTR. I had the privilege of coming on as a liaison to NCTR from CTP prior to taking this Branch Chief position and it's been nothing but a

pleasure, which is why I should end in thanking my colleagues who've helped to continue this collaboration on the very positive course it's been on.

I will take any questions if there are questions.

DR. LANZA: Are there any questions? We have a few minutes. If not, why don't we take this break just for maybe five minutes or ten minutes, rather than the previously planned break.

(Break)

Agenda Item: Center for Veterinary Medicine

DR. LANZA: What we'll do is we'll go through the next two talks from the Center of Veterinary Medicine and Office of Regulatory Affairs. And then if there is a little bit of a time, we can take a break. But at 3:40, which is when it should be actually 2:40, we're going to start the NCTR Division Directors' Overviews. And the goal is to be done at 4:55. So, if we can keep on track, we can be out of here at 5:00. So, the next talk will be the Center for Veterinarian Medicine, Dr. Whitehouse.

DR. WHITEHOUSE: Thank you very much. Good afternoon. So, our Center really is different than all the other Centers. So, at the Center for Veterinary Medicine, we approve new animal drugs. Also, apparently, some people

eat animals. So, we also ensure the safety of the foods that for human consumption. We're also responsible for the safety of animal feed. This includes feed that goes to domestic animals in addition to the food that you feed your pets.

At the Office of Research, where I work, we conduct research in a variety of fields: microbiology, residue chemistry, veterinary medicine, this includes research on certain biologics such as stem cell research and, also genetically engineered animals. We have the facilities to do animal research and we conduct research on cows, pigs, chickens, fish, frogs, and dogs.

Our office also houses two programs. One we call Vet Learn, which is a network of labs throughout the country and even some internationally where we investigate diseases of animals and outbreaks, also, mainly related to the products that we regulate. This is primarily related to animal feed and pet food. Our office also houses the NARMS program, which is the National Anti-microbial Resistance Monitoring System. And we do surveillance for antibiotic resistant bacteria throughout the world. And this is also through a network of laboratories, primarily state public health labs, but also some universities.

And I will also mention that the NARMS Program is an interagency program that's a collaboration with CDC and USDA and FDA, and recently this year we've also brought in EPA to make the NARMS program a truly one health surveillance program.

So, our collaborations with NCTR have been critical in helping us with our mission of protecting animal human health. Many of these collaborations fall into categories such as in vitro toxicity studies, nanotechnology work and a large number of projects related to microbiology.

Just going to run through a couple examples. We work with NCTR in in vitro genotoxicity studies, looking at the contribution of new animal drugs and looking at their toxicity and also potential carcinogenicity. Another one looking at nanoparticles and in vitro toxicity assays, including development of nanoparticle type positive controls.

As I mentioned, we have a lot of projects related to microbiology and antibiotic resistance. This is looking at long-term exposure to small amounts of antibiotics and the effect of that in the human gut. And this is we're trying to develop, as was mentioned earlier,

gut-on-a-chip technology also to take this to the next level. This is an important area for us.

A project that I've actually been involved with a little bit is to use a three-dimensional cell culture model to better evaluate virulence potential in mostly salmonella, but other bacteria as well. And this is basically using a combination of different cell types to develop more of a three-dimensional cell culture type system.

We're also working on looking at plasmids. We're heavily involved in plasmid encoded factors. These could be AMR genes or virulence factors. And Steve Foley in his group here are developing databases for plasmid encoded factors and we work closely with them. We have a PacBio where we can close the plasmid genomes. And also, very similar, Steve and his group are looking at developing a database for virulence factors of salmonella. And we collaborate with them using our large NARMS collection of salmonella to validate those virulence factors. We also look at studies looking at the intrinsic multi-drug efflux pumps in salmonella and their structure. Efflux pumps, as you know, are important anti-microbial factors in bacteria.

Demonstrated success of these collaborations. I was always taught never to use a font less than eight, but you're not supposed to read this. It just shows the number of publications that have come out of these collaborations. And also a few years back, there was an FDA award. This was a cross-center with our folks, CFSAN, and NCTR Scientific Achievement Award. This is for the melamine issue that was very important a few years ago.

I'm all ears and I'm open to questions. My e-mail address is there if you want to follow up with me for any other questions. But I'd be happy to take any questions right now as well. Thanks.

DR. LANZA: Thank you very much. Do we have questions for Dr. Whitehouse?

DR. KASPAR: Thanks, Chris, for your presentation. Is there a lot of crossover in the development of the database from your NARMS isolates and clinical isolates that CDC might have access to as far as gene markers for antimicrobial resistance?

DR. WHITEHOUSE: Everything that the CDC does with the NARMS isolates are sequenced and uploaded. So, we do have access to all those data and that's data that could be accessed by anybody, really. So, yes, they're all available. ??

DR. KASPAR: Oh, that's fantastic. I'd be interested in looking at that data. Thank you.

DR. WHITEHOUSE: Yes. Also, just to mention, we just started looking at seafood this year in our NARMS program. And also, just started a pilot project with the EPA looking at water samples from rivers and streams throughout the United States. And we expect to expand that over the years as well.

DR. LANZA: Anyone else? Well, thank you.

DR. LANZA: Thank you very much. And the next speaker is from Regulatory Affairs. Dr. Stromgren and she's the Director of the Research Coordination, Evaluation.

Agenda Item: Office of Regulatory Affairs

DR. STROMGREN: Thank you to the meeting organizers for this opportunity to present on ORA. And I'm happy to be here in person this time. At the last meeting in April, I joined via phone. So, I'm glad to be seeing all of you in person. I'm Selen Stromgren and I'm the Associate Director for the very new Office of Research Coordination, Evaluation, and Training. Only a little over two years old.

I appreciate that each of the Product Centers is different than the next and unique in what they regulate,

but ORA is really a different species than a Product Center. So, I'd like to spend a few minutes talking about what ORA is. Even its name starts with Office rather than Center. It's actually the second largest component of FDA after CDER. So over 5,000 employees and its nationally distributed workforce which could be challenging and exciting all at the same time. We even have some presence overseas at some of our international offices.

I've seen ORA being described as sort of the boots on the ground of FDA where rubber hits the road. About 75 percent of our workforce is comprised of inspectional employees. So, these are our consumer safety officers who every day go out the door, go inspect firms, collect samples to be tested by ORA's regulatory laboratories. ORA also is the component that carries out enforcement actions, working with the Centers, of course. We have an Office of Criminal Investigations. It's a very multifaceted mission. The core mission of ORA.

It is not a guidance setting, rulemaking, nor a peer approval component of FDA. Those actually are core functions of the Product Centers. However, I said 75 percent of the workforce is inspectional. All the rest, 25 percent is the laboratory workforce. Again, it's a nationally distributed laboratory network and these are

regulatory laboratories. And my colleague Dana from CTP gave a very nice definition of what regulatory science is. So, our main scientific mission is to support Agency preventive and enforcement action via our laboratory findings. Our Strategic Science Plan reflects long-term tactical goals to uphold this main mission.

So, I've listed some of the highlights of our Science Strategic Plan. Quality, and integrity of science, we have a lot of emphasis on this. It produces defensible results. It's very common for our laboratory findings conducted on FDA official samples to end up being discussed in a court of law. If the Agency enforcement action is challenged and we go to court, we have to be able to defend our analytical findings upon which the enforcement action was based.

Lab capacity with maximum efficiency. Again, our laboratory network, just like our consumer safety officers who go out the door every day, they receive samples through their doors every day, regulatory samples to be tested for various chemical or biological agents. We have to be able to perform this daily function efficiently with maximum capacity afforded by the number of employees we have. And we we've sort of resorted to thinking outside the box to enhance, expand this lab capacity. We've been

partnering with states. Sometimes they can provide a search capacity for us. But of course, in order for that to happen seamlessly, we have to establish some criteria for acceptability of state data for FDA regulatory purposes.

Lab capabilities. This is where our research function comes in. We're always on the lookout for new methods to support our regulatory function, how to analytically enforce FDA regulations on the product safety and efficacy of the products that the Agency regulates. And we need to be able to, while running fully validated methods, unofficial samples, we also need to develop methods that are investigative in nature because sometimes we don't know what's wrong with a product that comes in through the door and we have to run different tests to figure out.

Horizon scanning. Again, what new capabilities we need to develop for the next public health issue or for the next-generation products. We're always faced with new outbreaks or the possibility of new outbreaks, new contaminants, new products that present new risk attributes.

New risk perception. So new light can be on old products. In fact, PFAS, Suzy mentioned, perfluoroalkyl

substances, for instance, they're recently in the headlines because people perceive new risks associated with these substances. And new legislation, of course, we have to keep up with all this.

We have focus on producing data that can cross borders. We follow accreditation standards. We use standardized methods, user reference standards highly encouraged in our laboratories, and we conform to voluntary consensus standards. Again, this makes sort of international collaboration communication that much easier. Comparison of data. Use of other regulatory agencies' data and so forth for our intelligence purposes.

Timeliness. Speed and streamline decision making. These are important concepts for us as we develop our analytical approaches. Modernize technology base. Every year we invest a respectable amount of money in buying new equipment for our laboratories, establishing performance standards for laboratories whose work is subject to FDA review for regulatory decision making. Again, we talked about safe laboratories when they send data to us for our action, but also private laboratories that are usually hired by importers whose products may have been detained by FDA due to some nonconformance. And it is on the importer to test their future products, to

get their future products tested, to show that they have eliminated that contamination FDA found, so FDA can remove them from the detention list.

And in order to do that, importers hire private laboratories. It's a huge industry out there, but the work they do is subject to FDA review. In fact, the ORA Laboratory personnel spend quite a bit of time reviewing laboratory packages submitted by private labs in support of releasing various importers' products. So, we have to develop a performance standard, publish guidance for this industry to follow so we can accept their analytical packages.

And we're also involved in providing support and technical assessments to laboratories and national or foreign for purposes of lab capacity building. Several years ago, we had a pretty active lab capacity building program sort of going to countries that sort of have developing an infrastructure or they're trying to stand up some sort of regulatory frameworks like Costa Rica. And we went to those countries sort of assessing their conditions and coming up with suggestions for them what they could be looking for in the products they grow, they consume domestically, or they export out to other countries.

So, as I said, our research is quite different from Product Centers research in the sense that it's not as basic. It's very applied and it's designed specifically to support our regulatory testing program.

So, I mentioned are our laboratory network. We have 16 laboratory programs at 13 geographic locations shown here on the map. The laboratory programs are specialized along product lines. So, we have laboratories that test for food and feed products exclusively. So, they work on CFSAN issues and CVM issues. We have a laboratory in Atlanta that does tobacco testing. We have laboratory that specialize in device testing. Number of laboratories in pharmaceutical products testing and so forth. And this sort of alignment of the laboratories by product area was done recently in 2017.

So, this very applied research landscape and again, by research, ORA research really almost exclusively refers to method development. This is the breakdown of the various research projects that we have. As you can see, this is just the various areas fall under the different Product Centers' purview and they're color coded by the different laboratories. And don't worry about the acronyms, all our laboratories have sort of long acronyms.

But we work in areas - cosmetics testing, Suzy talked at length about cosmetics and challenges associated with those. So, we're involved with the tattoo ink projects as well. Development of applications for portable instrument platforms. This has always been an area of interest for ORA. So, we're always on the lookout to expand our laboratory testing capacity. One way to do it is to bring the testing to the points of entry, especially on the import product where most of our violations are. So instead of shipping all the products to the laboratories for fixed lab testing, perhaps we can develop methods that can be implemented at import points of entry, mail facilities, and so forth that can screen products for egregious contaminations. And then at least those can be detained right there. And that's the trickier or less obvious products can be shipped to the laboratories for the in-depth fixed lab testing. Field examination is another way of doing these things at the points of entry.

Forensic testing. And we have a lot of issues with counterfeit products, pharmaceuticals, tobacco products and so forth. Nanotechnology is in fact is a big area of intersection we have with NCTR. Our Arkansas Laboratory has a nanotechnology group and that's co-located on the same campus as NCTR.

Product assessment at preapproval stage in an area we mostly work with CDER on that. So CDER reviewers look at new product packages from sponsors, there are some claims there or some data there that they are not quite sure they'd like validated in the laboratory. And sometimes we'll do those kinds of testing for CDER.

Rapid Detection Technologies. Again, these are things that we'd like to be able to deploy during outbreaks, adverse events. The most recent one I can think about, several years ago there was a Deepwater Horizon oil rig explosion in the Gulf of Mexico. And we actually had to deploy - and this is performing a lot of fixed lab testing - we did deploy some mobile laboratories to the area, and we were trying to figure out how we can do some of that testing, looking for oil markers, oil contamination in seafood in a really rapid manner. And we evaluated some headspace gas chromatography and some electronic noses. So those are the kind of rapid technologies that we always evaluate, and they can be very helpful during an adverse event such as that one.

In tobacco testing, I've mentioned when CTP first was stood up, we actually worked with their Office of Science quite a bit. The first ban that they were responsible for enforcing was the flavor ban. So, we had a

lot of sessions with them trying to figure out how to stand up a method that was going to look for flavor compounds in cigarettes. At that time, their rule only covered cigarettes and so forth.

So, like the other Centers, it's important for us to track our research, especially given we're a geographically dispersed laboratory network with 16 different programs. We have to make sure we have a cohesive research program and all projects really are designed to support our regulatory mission, our regulatory testing core function. So, this is why my office, Office of Research Coordination, Evaluation, and Training, was stood up as part of the 2017 reorganization. So, there was no central research office prior to that that really oversaw the whole ORA research landscape. So, as a consequence, there were a lot of projects that were sort of being pursued because it was somebody's personal interest, or somebody thought that's the hot new thing to do. There wasn't really good alignment with the Centers or communication. There was duplication of effort and so forth.

So, my office, ORCET is sort of the acronym, is expected to manage the research portfolio and make sure it's designed in a way to benefit the Agency in the best

way possible. So, in order to do that, it's always tricky to sort of develop metrics to measure what we're gaining from the research conducted at our various organizations. We've sort of identified some impact factors and identified some tracked outcomes to monitor that, whether a given project is fulfilling that impact factor.

So, I'll just go through several of those. So, for this impact category number one, it's the bringing visibility to ORA science. Again, being not a Product Center, our research function is not always highly visible at the Agency level and beyond. So that's sort of one metric we look at whether a proposed project achieves in bringing visibility to the ORA science. And the tracked outcomes could be things like poster presentations, publications of laboratory information bulletin, publication of official methods, and of course, publication of peer-reviewed scientific articles. And in blue there on the left, I've listed sort of the larger initiatives, Agency-level ORA initiatives that the impact factor is associated with.

Our impact category number two is increasing the diversity of ORA portfolio. Some of the direct tracked outcomes are the, for instance, could be a first use of a new technology or an instrument, analysis of a product

never tested before, or starting new work which was new to ORA laboratories.

Our impact category number three, is increasing efficiency/confidence in the method or increasing applicability of a method. Some of the tracked outcomes will look at increasing the menu of analytes for a given method, expanding the matrix scope. Some food methods, for instance, can be only applicable to very specific things like apples and pears or a family, high water products and extending that to other matrices. Increasing throughput capabilities due to decreasing preparation or analysis time or conferring single lab validation or multi-lab validation status to a given method.

Again, being regulatory laboratories and our data can be discussed at a court at any time, it's very important that we always use fully validated and qualified methods. And sometimes this work validating a method, it is considered research for us. As you can imagine, there are not a lot of people always put in those types of proposals because people think that's not innovative work, new method development is, but validating methods is very critical for us.

Our impact category number four is valuable addition to analytical method preparedness toolbox. So

again, this sort of goes back to Horizon scanning. Have you developed a method that addresses a current FDA priority, that addresses an emerging issue of public concern? And hopefully if something hits us, we will be ready with a method that's already in our toolbox. In my like over now 11-year career with FDA, there have been quite a few times where we did not have a method ready to go and the emergency outbreak hit us. And as samples are raining at the door of our laboratories, our scientists are in there trying to develop the method at the mercy of science. A lot of pressure from everywhere. Deepwater Horizon again. At the time, we didn't have fixed-lab oil marker methods. And the Gulf states were calling the Agency all the time because they had suspended all fishing activities and that was very dramatically impacting their local economy. And then at FDA, we're trying to stand up a method with the right specs. So, it was a very stressful time. So, this is very important for us to be able to do intelligent Horizon scanning and have some methods in our toolbox ready to go.

Impact category number five, whether a method has been using in a coordinated agency response. So, ORA works with the Centers as sort of the crossroads for where all the Product Centers. We have a connection to each and

every one of them. And we provide surveillance testing for the various Product Centers. We perform compliance assignment, outbreak response, emergency response. So again, if our methods are used in any one of these to uphold the Agency mission, that's considered positive impact for that research.

So, we've actually gone to our scientists in the ORA laboratories and did a survey in terms of what they perceived was the impact of their work was. So, this is based on self-reported impact by the PIs. It's a word cloud representation where the largest font represents the most highly cited impact. So, by far, most of our research work apparently goes into supporting our compliance program testing. And this is again, testing official samples for agency enforcement action.

Last segment of the talk, I'll go over some of the scientific intersection points we have with NCTR. We have a lot of joint memberships on various Agency-level committees. And I've listed quite a number of them here. The Nano Task Force, that's probably our largest intersection point. Nanoplastics Interest Group. That's a relatively recent group and we've sort of been added to that group recently. Perinatal Health Center of Excellence. Dr. Slikker talked about that. Advanced

Manufacturing Technologies Work Group chaired by CDRH. Foods Program Regulatory Science Steering Committee, chaired by CFSAN. SOLAR, that stands for Sunscreen Operations, Laboratory Analysis, and Research, chaired by ORA. We have the FDA Mass Spectrometry Workgroup, chaired by ORA. The Emerging Sciences Council that was mentioned again. The AI Workgroup and the Senior Science Council.

And these are the current collaborations we have with NCTR. Again, in the interest of time, I'll just briefly mention the first one is a toxin bioassay. The determination of a human health hazard in regulatory food samples. NCTR of course having the animal facilities. Currently the gold standard for botulinum toxin is the mouse assay. People have been working and working trying to replace that with an in vitro assay, but it still remains the gold standard, still remains the regulatory standard. So, ORA works with NCTR to perform this test on samples where regulatory action may need to be taken.

Developing an intelligent recognition system for storage pest fragments contaminating food products. So, this was sort of started that there was an earlier project that was done as a proof of concept regarding use of this technique. It's an imaging technique for field elements or it has a large field program with CFSAN where we look at

insect fragments, glass shards, and metals in foods. And this is sort of an AI project that's trying to automate that. So instead of relying on microscope viewing by human, trying to train an AI system to recognize various field elements.

Third project is developing a novel data mining, data visualization method for safety surveillance of the FDA adverse event reporting system. And this has to do with, of course, FDA gets a lot of data from the public, from various intelligence sources. These are narrative reports and that's always been a challenge, how to automate searching those and coming up with patterns and trends that can serve as intelligence, that can direct your focus and research.

Design and development of machine learning algorithms to assist with automated pattern recognition of persistent organic pollutants in foods and feeds. This POPs program, persistent organic pollutants such as dioxins is a large program. Again, with CFSAN, we test a lot of samples every year looking at these biocumulative compounds and the data analysis associated with this is actually very time intensive. So, we're working with NCTR to automate that. And there's some preliminary data it can

cut down this from hours, done the traditional way, to only minutes.

And this last one is the Information Systems Management Project, the automated laboratory information or the LIMS. ORA has been working on this for several years now or more than that. Having again a large laboratory network producing data every day, there's always been a need to be able to share that data across the Agency. You know, various product Centers would like to look at the results associated with samples, products under their purview. At ORA headquarters we'd like to look at the data produced by the laboratories. So, this automated laboratory system was going to accomplish all that, but it's very challenging. We've looked at some off the shelf systems that had mixed success. So now we're working with NCTR to develop some custom laboratory information systems.

And I'll end with our areas of future potential collaboration with NCTR. We're looking forward to continuous partnership on initiatives such as the Nanoplastics, the PFAS, artificial intelligence, and collaboration on large data manipulation and trending tracking pattern recognition. NCTR has the expertise in

those areas. We probably have the data. So that's just a union that we'd like to keep facilitating.

Chemical signals intelligence exchange. This is important for Horizon scanning for ORA Laboratories, just compounds of interest, emerging health hazards, and of course assistance with toxicological assessment and health hazard evaluation of target compounds in ORA methods.

And so, as I've said earlier, ORA stood up as the first office dedicated to management of scientific research. So, it is an exciting time for ORA. We're now more active in establishing collaborations with the Centers and doing research that expands our knowledge base and capabilities. And a great example of that is the Perinatal Health Center of Excellence under the NCTR leadership. We were made to feel as a valuable member and it has been a launching pad for us, allowing the expansion of our research paradigm to fit with PHC interest. And by writing some proposals for this PHC, we've actually expanded our research into some novel territory.

This is a two-minute video. If it plays, we can watch it. It's not going to play there. But this was a two-minute video we had developed for ORA, especially focusing on this Office of Research. So, with that, I'd like to end and thank you for your attention.

DR. LANZA: Thank you very much. Do we have any questions? I just have one, if I may. And then we can take a short break and start. The question I have is the regulation of incoming drugs from foreign countries. You mentioned that there are regulatory parts of your program that are in foreign countries and I'm just wondering how you are able to regulate or maybe find fake drugs or drugs that aren't made to the standard that you expect. Or is that outside your world?

DR. STROMGREN: That's an area CDER usually sort of - they set the standards for the US, but it is an interesting area. There are a lot of conferences, in fact, between the European Medicines Agency and FDA, and even some other countries like Japan, India, they all have their slightly different regulations. So, we're trying to sort of align as best possible, but that does represent somewhat of a challenge in terms of if something gets approved in one country it's not necessarily approved here. There is no transferability of approval, if you will.

But I have attended some of those conferences where those issues were discussed. There is a concerted effort to align as best as possible, but some differences remain. Even on the food side, there are differences in,

for instance, pesticide tolerances. So, for instance, our domestic growers that export food out to European countries have to abide by their pesticide regulations. And usually, generally speaking, Europe has lower pesticide allowances in various products compared to us. So, when they export out, they have to abide by that country's rules and regulations and that could be different levels than what they can domestically distribute. So those differences exist. Various regulatory agencies are aware of it, but that's a good question, though.

DR. LANZA: No more questions. It's 2:36, so may I suggest you take five minutes, or almost. We'll start at 2:45 and then we're going to go and we're going to be done at five. Three talks, no more than 45 minutes each.

(Break)

Agenda Item: NCTR Division Directors: Overview of Research Activities

DR. LANZA: Okay, we have three talks. Maximally, it's 30 minutes presentation, 15 minutes of discussion. And our first is from the Division of Biochemical Toxicology, Dr. Gamboa da Costa.

Agenda Item: Division of Biochemical Toxicology

DR. GAMBOA DA COSTA: Thank you. Clearly, I'm not Fred Beland the Division Director. Fred apologizes for not being able to be here. And so, he asked me if I could give the presentation for the Division.

So, I think that all the presentations for the Divisions are structured in the same way so that there is more or less a continuity and it's easier to follow. The differences that characterize each Division, but an important element is always to give you an idea of the staffing of the Division.

So, we are currently composed Division of Biochemical Toxicology by 31 research scientist staff fellows and visiting scientists. We have nine support scientists and two administrators. On top of that, we currently have six ORISE postdocs and graduate students for a total of 48 staff members. So this is just a very slight reduction from the previous time that we gave you an overview of the Division and it stems from the reorganization where the chemistry support group that used to exist within the Division of Biochemical Toxicology was transferred to the Office of Scientific Coordination, and in return we incorporated the inhalation core facility into the DBT.

So, we keep collaborations essentially with all the Product Centers and with all the Divisions. We collaborate to the Division of Bioinformatics and Biostatistics, Genetic and Molecular Toxicology, Microbiology, Neurotoxicology, Systems Biology, and the Office of Scientific Coordination. With all the Product Centers: CBER, CDER, CDRH, CFSAN, CTP, and CVM. And although it's not explicitly stated here, we keep very tight collaborations and links with the Office of Regulatory Affairs, mainly at the level of technical cooperation and helping sort out issues, which goes both ways.

We keep very straight collaborations with NIEHS, Division of the NTP, and I will give examples of ongoing research that is being sponsored under the inter-agency agreement with the NIEHS NTP, with the NCI, the EPA, CDC and various universities within the US and outside of the US. From the standpoint of our global leadership and outreach, we are very much engaged in IARC. We often send scientists within our Division to participate in review exercises. The WHO, EFSA, OECD, and the Food Safety Commission of Japan.

The mission of the Division of Biochemical Toxicology, we have defined this to conduct fundamental

and applied research designed to define the biological mechanisms of action underlying the toxicity of FDA regulated products. And it has remained essentially unchanged for a long time. The problems remain the same. The way to tackle the problems is naturally evolving substantially. The specific goals of the Division are to characterize the toxicities and carcinogenic risks associated with chemicals, specifically those of interest to the FDA. And our strategies entail the conduction of bioassays, mechanistic studies, and computational modeling. And this is really just a very bird's eye view of what we do. What we do is actually quite broad in scope.

To give you an idea of the top three accomplishments that we have identified for the Division in 2019, I'd like to start with essentially the finalization of the core study of CLARITY-BPA consortium, which was crystallized initially in an NTP research report. And more recently, we have published the outcome of the course study in a publication by Camacho et al. in Food and Chemical Toxicology.

We would also like to highlight a series of studies conducted by Dr. Doerge on the pharmacokinetics of

arsenic. And I will be going a little bit more detail on this work and the proposed work that we are considering.

Finally, we would also like to highlight the work conducted by Igor Pogribny's group, which address the epigenetic mechanisms that may be able to justify organ-specific carcinogenicity of acrylamide. It's really interesting, but in a nutshell, acrylamide is a known genotoxicant, but the genotoxicity alone is perceived by mapping DNA adducts across a range of organs but does not justify the organ-specific carcinogenicity that we have observed. And so, this study sheds some clues as to why cancer is developing in some organs and not in others.

Now, moving into representative current projects I'm going to be presenting. To the first one is the work on Tier 2 pigments that Dr. Fitzpatrick has alluded to. So, this work is being conducted in collaboration with CFSAN, and it's being sponsored by the Perinatal Health Center of Excellence. So, adults between the ages of 25 and 39 years of age have the highest tattoo prevalence, which reaches up to 55 percent. And importantly, women of childbearing age have higher rates of tattoos than men.

Very interestingly, the average tattoo contains 250 milligrams of tattoo pigment, which was something that surprised me. And as Dr. Fitzpatrick indicated the origin

of the tattoo dyes is not necessarily designed to be put into human skin. And 30 percent of the US population has over four tattoos. So, this really goes to show the burden of dye people are exposed to.

Very interestingly, upon having been tattooed, most of the pigment tends to disappear from the site of the application. So, between 87 and 99 percent of the pigment has been reported to disappear. So, no one really knows exactly where all the mass transfer occurs. So, we know that some of it ends up in the lymph nodes, but no one really knows the final fate of the pigments. And so, since the highest rate of tattooing is found in women of childbearing age, there is concern about potential exposure of the unborn fetus to the tattoo pigments that may occur via placental transfer.

So, the hypothesis that we put forward in designing this study was that the intradermal injection of tattoo pigments into the dorsal skin of pregnant mice might biodistribute to the organs of the dam and possibly to the developing fetus via placental transfer. So the experimental design that we put in motion was aimed to assess the placental transfer and biodistribution of three commonly used azo tattoo dyes: pigment orange 13 in the left, pigment yellow 83 in the middle, and pigment red 22

on the right. So, at this stage the status of the protocol is that we need to have radio-labeled tattoo dyes, and this is easier said than done. But I think that we're finally getting close to the point where we may be able to obtain these dyes. And actually, I would be remiss if I did not mention that all of this work is being coordinated by Dr. Boudreau, who's the principal investigator in this study, in collaboration with CFSAN colleagues.

So, once we get radio-labeled tattoo dyes, we're going to tattoo SKH-1 mice at the rate of 2.5 milligrams per square centimeter with the corresponding amount of 10 millicuries per mouse. And then we're going to evaluate all the radioactivity distributes in the fetuses, in the organs of the dam, and the excrements, to get a proper assessment of how the dye distributes.

We're also going to try to ascertain, by comparing the outcome in pregnant and non-pregnant dams to see if pregnancy influences the biodistribution of the dyes into dams. So, we hope to be able to give you updates on these studies in forthcoming SABs.

Okay, for a second representative current project, I would like to highlight the work that we are conducting on pegylated biopharmaceuticals. And again, in the CDER presentation, there was an allusion to this

collaboration. This work is being conducted by Jia-Long Fang, the principal investigator in this. And it's an example of work that is being sponsored by an inter-agency agreement with the NIEHS division of the NTP.

So PEGylation is the process of both covalently and noncovalently bound PEG serves to essentially mask drug proteins, which improves the solubility of these drugs, extends the circulating half-life, increases drug stability, provides and add protection from proteolytic degradation, and ultimately can reduce dosage frequency without diminished efficacy and with potentially reduced toxicity.

So, the issue is that there is that indicating that several PEGylated biopharmaceuticals have caused PEG accumulation and cellular vacuolization in a number of tissues, particularly worryingly in the choroid plexus. So, this is the outcome of pre-clinical studies and it alerted the FDA to lack of data in this field.

There is also enhanced concern about these in certain populations, namely pediatric populations, or populations that rather than just having a short round of treatment with biologics, actually require lifelong exposure to these biologics. So, there is lacking data about how the tissue levels of PEG vary over time in

prolonged exposures, and about the toxicology of PEG on some tissue, especially in the choroid plexus and the kidney.

And this is a study that is being conducted in collaboration, both with CBER and CDER. And although I cannot give you the specifics, to be honest, there is biologics that are regulated by CDER, others by CBER.

The experimental design was to assess the toxicity resulting from weakly repeated subcutaneous or intravenous injections of high-molecular-weight PEGs, and these were 20, 40, and 60 kDa for 24 weeks in Sprague-Dawley rats. So, this is the toxicological component of the study.

And then we have the two pharmacokinetic or toxicokinetic studies. One entailing a single dose. So, to evaluate the toxicokinetic profile of high-molecular-weight PEGs, even as a single subcutaneous or intravenous dose to Sprague-Dawley rats, followed by repeated dosing pharmacokinetic assays to try to ascertain or simulate better conditions that humans are exposed to.

And so, the tox study is now the end life stage has been completed. The pathology is being conducted. And again, we hope that in the next update we should be able to give you a better idea of the findings. However, we

have some preliminary data that we can share with you and which pertains to the levels of PEGs in the plasma, urine, and feces of these rats from the tox study. And it's a quite dense and you probably can see the molecular weights. But essentially, we have in the x axis, we have the data for 20 kDa subcutaneous, 40 kDa subcutaneous, 60 kDa subcutaneous, and also 60 kDa IV. The reason why we're doing that, specifically the IV and subcutaneous for kDa is to try to ascertain whether the route of administration bears any impact on the pharmacokinetics.

So, in essence, what you can see is that the serum levels increase with the molecular weight of the PEG, and this is in the plasma. And that in the urine, you get excretion, you get much more efficient excretion of the low-molecular-weight PEGs than the high-molecular-weight PEGs, which justifies the concentrations in plasma. On the feces, there is really not that much of a difference. So, in a sense, this was known to a certain extent. I am not entirely sure if anyone had actually compared across these three molecular weights. So again, this is how the data stands. We hope to have more data soon which we share immediately with the Product Centers to keep everyone apprised of how things are moving along.

Now, talking about future projects, I'm going to be talking to you about two future projects. The first one, it's an arsenic bioassay. It's still essentially in discussions with the CFSAN. The second one that I'm going to be talking to you about is already undergoing internal review, initial internal review at NCTR.

So average arsenic concentrations in drinking water in the US are approximately two parts per billion only. But some areas have concentrations that can be as high as 1,000 parts per billion. That's one part per million. The EPA maximum contaminant level in WHO guidelines for this inorganic arsenic in drinking water is only 10 parts per billion. So, there is a population that is being exposed to levels that are substantially higher than the maximum recommended EPA concentration.

And considering the US population, the estimated daily exposure to inorganic arsenic is between 0.08 micrograms per kilogram body weight a day to 0.2 in adults. And then as you go to children the exposure gets higher. So, in children between ages of one and six, it's slightly higher between 0.11 and 0.32 micrograms per kilogram of body weight a day. When you go to children under one, because of the ratio of diet that they consume per body weight, and also probably because of the content

of arsenic in rice that is used for baby formula, it's substantially higher, between 0.24 and 1.19 micrograms kilogram body weight per day.

So, as I indicated previously, we have the DBT Dr. Daniel Doerge conducted the very extensive assessment of the pharmacokinetics of inorganic arsenic and also organic arsenic. And again, this was conducted under the interagency agreement with the National Toxicology Program. So, these are the key points. It's a wealth of data. It's many, many papers that were published on these in the last couple of years, but these are the key points. Inorganic arsenic is readily absorbed by the GI tract and so is dimethyl arsenic 5. The metabolism is dominated by dimethyl arsenic 5. And very importantly - and this was not really known before - the toxicokinetics for arsenic and are non-linear above an exposure of 50 micrograms per kilogram of body weight a day. This has implications to be considered when you consider that from high exposure studies.

DMA 5 can be reduced to DMA 3, which can react with (indiscernible word) in proteins. So, there is also an element that now we understand much better all the cycling of the arsenic species. And now we have reasons to believe that the dimethylarsinic 5 may also prove to be a

toxicant, which is novel data. Also, importantly, and this sets the stage as to why we believe that new studies are warranted, there is very poor lactational transfer of arsenic species. Meaning that if you conduct a study where you only treat the dams, the pups are really not being exposed to arsenic or to any appreciable amounts.

So, if we could summarize the outstanding data gaps on inorganic arsenic. So, there has been a number of studies that have been conducted and notably two studies conducted by scientists currently at the NIEHS. There is an unusual dose response for lung tumors. That suggests non-monotonicity, but that's when you consider they aggregated the curve from two separate studies. When you consider those two studies, there is also inconsistencies between the target organs across the two studies.

And finally, t although those studies were called lifelong exposures to arsenic, they did not really encompass appropriately the early period of development, which may be a critical one for exposure to arsenic. And then finally, with the dimethylarsinic 5, again, there are studies and it is known to be a rodent carcinogen. The studies have indicated so. However, there was no - due to the poor lactational transfer, there was really no exposure of the pups to the dimethylarsinic 5. And so,

again, an important element has been missed, which is the perinatal exposure.

So, what we are proposing is, in essence, truly a whole life exposure bioassay. And the idea would be to treat dams and sires before and during breeding, and the dams during pregnancy. Then the pups would be gavaged directly from post-natal day 1 to 21. And that's something that the NCTR has the expertise to do on a routine basis. And then upon weaning, the animals would keep on being exposures to our arsenic through their drinking water. And the idea would be to have two arms of the study, one, treating the animals with inorganic arsenic and another one with dimethylarsenic 5 to try to ascertain each of those responses and ascertain the potency, relative potency for the two species.

This is something that may not be entirely apparent all the time, but we very rarely at NCTR do we do just the standard bioassay. Typically, we encompass a number of other end points in an attempt to enrich and to get the most of that assay. And so, the proposal here would be to include the elements of internal dosimetry and ascertain epigenetic alterations on the animals, microbiome alterations, and also ascertain hotspot cancer

driver mutation in samples from the animals. And again, so this study is still under discussion with the CFSAN.

For future project number two, again, this one has already entered the review pipeline at NCTR. It deals with nonalcoholic fatty liver disease. I apologize. The previous study on arsenic is being put together by Dr. Camacho and Dr. Beland. This current protocol, the nonalcoholic fatty liver disease protocol is being led by Dr. Igor Pogribny. And the nonalcoholic fatty liver disease is the most prevalent form of chronic liver disease in the United States. And there's pronounced sex difference in human susceptibility to NAFLD, with the women being more susceptible to the disease than men. There is also extensive individual variability in the susceptibility to the disease. And there is a lot of difficulty in getting an early diagnosis done and staging the diseases. And moreover, there is no currently FDA-approved therapies for the disease.

So, the objective of this study is to determine genomic and genetic determinants of the sex and individual susceptibility to NAFLD and also hopefully to develop and evaluate novel biomarkers for NAFLD diagnosis and monitoring. And so this protocol is predicated on previous studies that were conducted at the Division by Dr.

Pogribny' Group and which entailed treating groups of three males and three females from 25 collaborative cross mouse strains with either a controlled diet or a high fat and high sucrose diet, which probably mimics fairly well the current diet in North America.

And so, the animals were treated for 12 weeks and they were sacrificed. And then out of all those 25 strains, two were selected, strain CC041 and CC042, because those were the strains that showed more of a difference beyond now. Males and females responded to the high fat, high sucrose diet. So, it's very apparent here that males seem to be substantially more sensitive than the females to the diet challenge.

So, what we are proposing at this stage and which is undergoing current review, is to select using those strains males and females, to put them again through a controlled diet, high fat, high sucrose diet. But this time take them for a longer time. So the intent is to start with groups of 30 males and 30 females and sacrifice one third at week 24, another third at week 36, and the final third at week 48 of exposure, so that we can also ascertain a time component of how the disease progresses.

The endpoints, we can see there are obviously histopathology, which is a crucial element, but also

clinical biochemistry, transcriptome analysis, epigenomic analysis, metabolomic analysis, and microbiome analysis. So, we hope to get this protocol approved and to initiate the studies as soon as possible.

I'm also showing here future project number three, which is really not a project in itself, but it it's more to highlight the fact that the CTP, the Joint CTP/NCTR Inhalation Toxicology Core Facility has been transferred from the Office of Scientific Coordination to the Division of Biochemical Toxicology. And the idea was to try to create more of a synergy with the expertise in toxicology that exists in the Division and the ability to conduct these studies. So, this was recent, and I wanted to highlight that change.

I won't really be going in detail in terms of the studies at the Inhalation Core has done because Dr. van Bemmel has given a very nice overview of the work that has been conducted and that may be waiting as in the future.

So finally, I would like to highlight some challenges that the Division is undergoing. And I do recognize that there is not much that the SAB members may be able to do about some of these elements. But I also believe that it is important for you to have a global

perspective of how the Division stands. As Dr. Heflich raised this morning, we are facing an increasing challenge in the recruitment of post-doctoral fellows and visiting scientists. The reason being that we are now precluded from recruiting anyone that has not been in the US for at least three out of the last five years, so that a proper security clearance can be conducted.

And I understand the reasons behind this, but I can also not, not take into consideration the impact that this will have. There is an immediate impact and there is a long-term impact. And what I can tell you is that a good number of the people that are present in the room, including myself, would not be here today if these measures have been implemented a long time ago.

So, the National Toxicology Program is under new leadership and the new leadership has defined a new vision and strategy that is entailing a modification of how we interact through our interagency agreement. The type of investment that is being made and the type of studies that will probably be coming in the future do not necessarily fit as well, perhaps as they did before in the Division of Biochemical Toxicology. And so, whilst other divisions may benefit from this change, we have to recognize the fact

that for the Division of Biochemical Toxicology, this may be a challenge.

And finally, and this is really not a challenge, and again, it's more to highlight that we're still in the process of integrating the Inhalation Core into the Division, but I'm certain that things will run smoothly and without any major issues.

So, with this, I finished my presentation and hopefully I'm still within my allocated time. Thank you.

DR. LANZA: Thank you very much. Right on time. Are there questions? Yes.

DR. STICE: I understand the importance and the value of the future project on nonalcoholic fatty liver disease. I guess what I'm grappling with a little bit is how does the objectives of determining the genetic determinants in the biomarkers fit in to your particular mission within the Division? If I look at your mission statement and then the objectives of that study, I guess I'm trying to get a better understanding of the mesh and how they fit together.

DR. GAMBOA DA COSTA: Yes, exactly. Well, so you have a perfectly reasonable question. I think it stems from the fact that when you try to typify in a sentence what is it, what the mission of the particular entity is,

you tend to narrow it to the point where it no longer reflects. So traditionally, we have a component that addresses directly immediate needs from the Product Centers when it comes to evaluating direct toxicities. But we also have a research component that is broader and that has historically given us a better understanding of the techniques, approaches. And I'm afraid that I don't think that Dr. Pogribny is here.

I don't think that I can give you a more specific question, so I think that it stems from the fact that we're not addressing the toxicology of an agent Right? Is that correct? Yes. So, I think that it just stems from the definition that we put forward in these slides, that it's perhaps a little bit overly narrow for the scope of the work that is conducted by the Division.

DR. GANEY: I have a related question about the nonalcoholic fatty liver disease project. You mentioned that there's no therapy for this condition. So how does it help the patient to know that they have the disease and how bad it is or whether it's getting better or worse? How does that help a patient?

DR. GAMBOA DA COSTA: So, one of the key challenges is diagnosis. It's very difficult to diagnose the nonalcoholic fatty liver disease. So, it's one of the

objectives of these studies to try to understand whether there are any perceivable changes in the - and again, bear in mind, I'm not the P.I. of this protocol. So this is my vision for this study is to try to understand if there is any changes in, say, for example, in the blood chemistry or in any elements that may be perceived that would allow us to catch the disease earlier on. So, it's about the possibility of intervening earlier sooner than later. So that's one of the elements.

DR. GANEY: Okay. I'm not sure that was really an answer to my question, but I'll let it go at that. Can I ask another question, a different question? To go back to your current project on the tattoo pigments, which I think is actually really interesting. You selected three pigments.

DR. GAMBOA DA COSTA: Yes.

DR. GANEY: And were they selected because of how much is in the tattoo ink or because you know there's some toxicity associated with them or for some are easy to detect analytically or how exactly were they chosen?

DR. GAMBOA DA COSTA: No, absolutely. Fair question. Okay. I can start with easy one, which is that from an analytical standpoint now, these are all terrible things to work with. They're designed not to be soluble.

They're very difficult to synthesize to radio label. There is nothing good about them. So, I would actually defer to Dr. Boudreau to give us a better perspective. Mary, would you mind?

DR. BOUDREAU: The reason the azo pigments were selected is because they are more commonly used right now because of the bright colors that they are able to produce. You see more tattoos with reds, yellows, greens, and blues that are bright colors. And most of these are your azo pigments and these are common pigments.

DR. GANEY: So, then my presumption is that if you find that they distribute to the fetus, then you'll follow up with toxicology studies.

DR. BOUDREAU: Yes.

DR. GANEY: Okay. All right. Thank you.

DR. BOUDREAU: This is pretty much of a preliminary study just to see if they biodistribute and to see if pregnant vs. non pregnant female mice - if the biodistribution differs between them.

DR. COSENZA: I have some questions on the pegylated studies. It's my understanding from reading this and what you said there's no protein attached to these PEGs. Is that correct? It's just the PEG is being dosed?

DR. GAMBOA DA COSTA: Yes. So, the initial intent was to start - when we started discussing this program with CDER and CBER, we thought about selecting an archetypical biologic. The first thing that we concluded is that does not exist. They're all different. And then until we could understand the basic toxicology of PEG itself that we did not want to tackle more complicated issues that could stem from immunological responses to the protein and not necessarily to the PEG. So, this is a very good question.

But right now, the toxicology of PEG is well understood and there is essentially nothing that we should be concerned about that I'm aware of. But it is really not known once it gets into the body. We know the site eventually the biologic is degraded. There is proteolysis and the remnant is PEG. So, we thought that this was a good way to start the studies.

DR. COSENZA: Okay. I would just say that my experience, which was on a number of pegylated proteins, where the PEG goes is very much dependent on the protein it's attached to.

DR. GAMBOA DA COSTA: It is possible.

DR. COSENZA: It may be somewhat artificial where these actually go not being conjugated, that's all.

DR. GAMBOA DA COSTA: So, to a large extent, what we've been dealing with is with really complicated challenges of method development. And so, the learnings from these protocols are going to make it much easier than to follow through with an actual test article that resembles more what people are exposed to.

DR. COSENZA: Okay, great. Thanks.

DR. LANZA: If I can follow up on that question. So, when I was looking at the data, one observation I have is that as you go up, it does appear all of them are large PEGs. As you started getting 40 to 60, I think what you're forming are micelles. And the PEG is now not just dispersed but gradually into particles. And one of the things that you see in that case is that it looked like you are overcoming the hydrodynamic size that you would filter through the kidney, which would be 6 nanometers, 7 nanometers, right? But in rodents, mice and rats, the situation, unlike humans, is that those PEG particles are going to go through the hepatobiliary system directly into the gut. And I think that's what you're seeing.

And of course, this is, as you say, without the immunological fact that these mice have never been exposed to PEG, while people, women in particular with cosmetics are. And that, of course, also changes it. So, you might

want to think about this. And I think Dr. Patrie(?) can help in terms of whether it's forming micelles and going through that phase.

DR. GAMBOA DA COSTA: Yes. So, I'm not entirely sure if PEG is given already as a dispersion in water, whether it will micellize or not. It's important information to consider. Thank you.

DR. LANZA: From biochemical to biophysical, and that's the big point there.

DR. GAMBOA DA COSTA: And when you start dealing with the - so for many, many years, everyone in toxicology was dealing with small things. The good PIH is nitriles. Once you start getting into the realm of something that weighs 60 kDa, things start being different. And so, yes, that's a good point.

DR. LANZA: Ken.

DR. RAMOS: Very enjoyable presentation. A quick question for you is, why is arsenic a focus of your program? How does that relate to FDA regulated products?

DR. GAMBOA DA COSTA: It's in food.

DR. RAMOS: And so, given the fact that it's oral exposure in food, is there a plan in place to follow up on that?

DR. GAMBOA DA COSTA: Yes. The pharmacokinetics studies are being concluded now and we are now entailing in a discussion with CFSAN on the next step forward, because this is, food obviously falls under the purview of CFSAN. And so, we're initiating a dialog to try to understand which are the appropriate next steps.

(Off mic comment)

DR. GAMBOA DA COSTA: The novelty on the pharmacokinetics comes from the fact that opposite from what was thought, organic arsenic may also be relevant.

DR. LANZA: Michael.

DR. SAUER: Quick question around the PEG and a little bit of information. We used, basically it was a three-armed 60 kDa PEG. And what we were doing was we were conjugating with a small molecule. We ran both 90-day as well as six-month studies in rat and dog. We saw quite a bit of tissue vacuolization, so that actually points to the idea of the fact that it's forming a particulate, probably being taking up by histiocytes. We never published that data. It's all been submitted to an IND. I think that might give you a lot of good information to be able to utilize, because what we also did in those studies was, we had it where it was the pegylated compound without the small molecules.

The other piece was, we've also done mass balance studies. I don't know if you're planning on doing that as well. It's a little more difficult with the linear PEG because it's going to get chewed up. And of course, you can't have one C14 or whatever type radiolabel per molecule. So, it's a little bit harder to do. We also did electron microscopy. So that's why we knew it was vacuolization in these different tissues. So that information may be able to help guide you. I can give you the IND number. The company's gone. The IND has been discontinued.

DR. GAMBOA DA COSTA: These things happen. Thank you very much. Any data helps because this is, as you're saying, this is very difficult to tackle. One of the issues that we had was obtaining radiolabeled compound. I think that unless we synthesize it in-house, it's difficult. It's easy to take one of the extremes by using methyl iodide and extend the chain a little bit, but getting the whole thing labeled is not trivial. And so, thanks. That's very useful. Thank you.

DR. LANZA: Any more questions? If not, thank you very much.

DR. GAMBOA DA COSTA: Thank you.

DR. LANZA: Our next speaker is from the Division of Bioinformatics and Biostatistics, Dr. Tong.

Agenda Item: Division of Bioinformatics and Biostatistics

DR. TONG: First of all, our Division is different from any other Division at NCTR. And so, we only do the dry lab work. And so, every other Division do the wet lab. And second, we have more supportive scientists compared to the research scientists in other Divisions; normally it is the other way around. However, we also have a lot of the vacancy in our Division. And this has been persistent for several years now. So, we normally run an 80 percent at full capacity for this Division.

At this point, we have a little bit over 50 peoples spread across the four branches in the Bioinformatics Branch, Biostatistics Branch, R2R, which stands for research-to-review and the return. And the last one is Scientific Computing Branch. And roughly speaking, 40 percent of the Division activity is the folks on research and 60 percent is in support and service.

So, in terms of the research, we mainly focus on five areas of the research. And are we doing a lot of the work in the endocrine disruptors. On this project that was established back into the 1996, way before this Division

was established and minimally we used the chemo informatics and the molecular modeling tools to develop a predictive models for the industrial chemicals, environmental chemicals, as well as the food additives and cosmetic compounds that are regulated by FDA. So, in nature, we have a lot of collaborations with the EPA and FDA and other stakeholders.

So, our Division is doing a lot of the work in the area of the genomics. And more specifically, we run a long-standing consortium activity since 2005 and this project already being published over 30 papers. Right now, we are around the fourth phase of this project to more focus on the precision medicine. I do have one slide to provide a little bit of an update on where we are in terms of this consortium activity.

We are also working with the rare disease, as you were aware, there are over 7,000 rare disease, but only 600 of them have the treatment options. So, what we do in our Division is to use the bioinformatics approach to systematically survey the marketed drugs. That means these drugs are already safe, on the market, and to see whether we can repurpose these drugs for the treatment of the rare disease.

And in terms of the drug safety, we mainly focus on the drug-induced liver injury. Now, as a Division mainly focus on the bioinformatics and the biostatistics. And naturally we use a lot of tools related to the artificial intelligence and machine learnings. And normally I give a presentation, I don't single out this particular activity. Now it's such a hot topic, so later on, I'm going to provide some of the update on some of the projects we are working in the artificial intelligence.

So, in terms of the missions and the research in our Division is to conduct integrated bioinformatics and biostatistics research to support FDA submission of improving the safety and efficacy of FDA regulated products. And our Division doing a lot of the support. And not only within the NCTR, but also collaborating with CDER and ORA. You already heard of some of the project that was mentioned by the representative from other Centers.

So, the mission for the support component of this Division is to ensure that the Division's activity related to FDA's review process. Our linkage with the Product Centers continues to be strengthened and our capabilities evolve to meet the current and the future needs of FDA.

So, the next few slides, I just give you a quick update about our support project. And our Division closely work with the on-site OMIT staff to taking care of the I.T. infrastructure and the related support activities. And basically, our Division function as a computer center to taking care of a little over 130 servers and the petabyte and the data storage.

Several years ago, we established a specific function in our Division to provide a bioinformatics data analysis support by establishing data analysis environment to manage a commercial and in-house software tools as well as conduct the training course. I just want to point it out that the training course was collaborative with the Office of the Scientific Coordination, and we provided a hands-on training as well as provided lectures on the basic principles behind the tools we have available at NCTR.

So, this slide summarizes some of the projects we are working on in collaboration within other Division at NCTR. Now, this is not meant to be exhaustive and it just gives you a glimpse on what type of techniques or expertise is mainly sought at NCTR. So if you look at the collaborative project list here, most of them all related to the sequencing data analysis and genomics data

analysis, and we do have two on text mining project with the Division of the Biochemical Toxicology, are related to the monograph program.

In this program the scientists literally need to look like 10,000 literature, narrowed down to hundreds to reference, using these references to generate at the monograph. And this is time-consuming and labor-intensive. So, we are working with them to using the text mining tool to quickly identify the relevant reference for the monograph generation.

And Dr. Lal-Nag already mentioned about FDALabel, and I'm not going to elaborate about this particular project. I just want to make a few quick notes on the FDALabel is to use to manage FDA drug labeling data, extrapolating data first. It's long. It's about 20 pages with a little bit over 80 sections. And each section was focused on specific information. So, this information is extremely useful for in the review process, particularly in the CDER.

Second, track labeling document, it's very big. And we have a little bit over 100,000 documents. So, during the review process the reviewer will have a very difficult time to identify or to find that the relevant drug labeling document. So, FDALabel is the database and

the manager of this labeling document with the user-friendly interface so the reviewer can get in, quickly identify the information they needed in the review process.

This project was led by Office of the Scientific Coordination at NCTR, with the expert consultant from the Division of the Systems Biology. And my Division just provide the muscles and do the heavy lift to develop the software. And this is the truly collaborative project that's not only within the NCTR, but also with the other Centers. For example, in CDER was led by an Office of the Computational Science. They collecting the requirement from the reviewers, provide the trainings, and the user support. This is the web link to lead to their website to describe their role in this project.

We also have a fantastic team from the Office of New Drugs and this team called a Drug Labeling Teams. So, they have a tremendous experience in use to drug labeling document to support the review process. They know that what information is supposed to looking for. They provided this input to be implemented in our software. So, the reviewer coming in can easily navigate through the drug labeling and database to find the information they need.

And also. And we have been communicating with the Office of the Generic Drug, as well as the Office of the Pharmaceutical Quality. And we also are in discussion with the CBER and the CVM. And the most other interaction was generated in this setting, and particularly in the last two years when we present FDA drug labeling and database and some of the representatives from the different Centers think that this is a great tool and really useful. So, they come back and make the connection to the people who might benefit from this tool. So since then, we have a lot of the communication. And we also made this tool publicly available through the Amazon Cloud.

In the past a few years, we are enjoying tremendously to collaborate with the CDER. And the number of the project already be mentioned by Dr. Lal-Nag. In the last year we completed the Breakthrough Therapy Designation systems. We also finished the text mining study of the regulatory document. And both projects are in collaboration with the Office of the New Drug. And right after I submitted this slide set and we just closed another project is a Risk Evaluation and Mitigation Strategy. This is the collaboration the CDER Office of the Communication.

Both DASH and IND Smart Template are already mentioned by others, so I'm not going to elaborate on it here. And I would just want to point it out, several months ago, we just had another new project with the CDER. This is called the Safety Policy Research Team, called the SPRT Team. They want us to develop a system to collect the information and to analyze the post-market safety actions, policy, and outcomes. And they already have some of the information available in the existing database in a spreadsheet. They hope we will be able to use these systems to bring this information, to connect this information with the DASH. And on top of that, they ask us to develop the Natural Language Processing tools to extract the information from the regulatory document to populate the SPRT system.

So, we also very fortunate to work with the ORA in the past several years. And Dr. Stromgren already mentioned all of these projects. And I just make one quick point. Arkansas Regional Lab is one of the labs in ORA located at NCTR and Dr. Stromgren already mentioned. And we really take full advantage because every day we run the van pool with them. And in and out for two hours every day, you got to talk about something. So, this is the discussion what came out from it. And the two projects are

related to artificial intelligence. One is for the storage of pest fragment identifications for the food contaminations. Another one is using the machine learning method to analyze the mass spectrometry data. And this is actually is a Chief Scientist Challenger Grant and the PI is from the ORA.

So, this word cloud representation for the 20 papers we collected in the past few years presenting the main activity in this Division. And then you can see the genomics is everywhere, including the RNAseq. RNAseq and genomics, next-generation sequencing. Even the reproducibility as related to the genomics. So, you can see this is one of the areas it was focus in our division. And we also, you can see the drug-induced liver injury also popped up. And the third one is big data and artificial intelligence.

So, in the next few slides, when I talk about the research I'm going to follow in this order. I'm going to talk about genomics, talk about drug safety related to DILI, and talk about artificial intelligence.

In terms of the genomics, as I mentioned at the very beginning, we have a long-standing consortium activity called the MicroArray and the Sequencing Quality Control. We started this project back in 2005. By 2014, we

completed the three projects. In all these three projects and we try to understand how we can come up with standards to assess the technical reliability and the clinical utility of the emerging technology, particularly genomics technology.

So right now, we are focused on the fourth project called Sequencing Quality Control Phase 2, or SEQC2, which is a specifically address the reliability and reproducibility issues in the area of the precision medicine using the next-generation sequencing. And for the first project after is the Working Group Number One, is focused on the cancer genomics using the whole genome sequencing. We already have one paper accepted by Nature Biotechnology and a second a paper in Nature Biotechnology under the review. And both should be out very soon. And we have additional two paper are going to be submitted to the Nature Biotechnology in the next a few months.

And second, the Working Group is focused on the cancer genomics, but are using targeted gene sequencing. Now targeted gene sequencing represented the current clinical practice using the next-generation sequencing and a whole genome sequencing actually only represent the future application of the clinical setting using the next-generation sequencing. So, we just got off the phone with

the Nature Biotechnology four paper thing to be submitted very soon. And Area Number Three is a focus on the reproducibility of the whole genome sequencing and we are scheduled to submit a two paper in FY20. And area number four is a focus on epigenetics and the epigenomics.

In terms of the drug-induced liver injury, I'm not going to elaborate on why we needed to study the DILI. One, urgent issues to the FDA as well as to the drug development or pharmaceutical company is up to 50 percent of the drugs failed in the clinical setting due to the DILI are not detected by existing preclinical models. That means the existing preclinical model cannot do the job accurately or correctly to predict liver injury in human. So, in the research community, there is a lot of the efforts using alternative methods, including Susan mentioned about using the liver-on-the-chip to assess or to replace some of the animal studies to assess the DILI.

And normally, some of these tox technologies are high throughput in nature. That means you're really able to screen in large number of the drugs. So, in order to do so, you need to have a large number of the drugs with the known DILI outcome in humans. So, this actually is a one of the focus in the project that we are doing for many years now called the Liver Toxicity Knowledge Base. And in

2011 we published the first list of the drugs. At that time, it was only 280 drugs with the DILI classification using the FDA drug labeling document. And five years later we published a second list called a DILI Rank, and a little over 700 drugs were annotated with the causality assessment. And a couple months ago we just published the third list called the DILIst. The DILI severity and the toxicity and we annotated around 1,300 drugs. And this drug list is going to be extremely useful to assess the reliability of the alternative methodology.

Now I'm going to shift gears a little bit and to talk about the big data analytics and artificial intelligence. And. AI is just a general concept and many times people talk about AI at the same times talk about machine learning, but actually machine learning just a bigger portion of the AI And within the machine learning that it also probably the most exciting methodology is the deep learning.

So, what I'm going to do, I'm going to talk about one particular project we are working on and the work the CDER called a DeepReviewer, which is not using the machine learning, but it's a part of the AI And also going to talk about the second project about genomics biomarkers for DILI and using the machine learning method.

And lastly, I'm going to briefly mention about the collaborative efforts with the PrecisionFDA. And this is belonging to the Office of the Chief Scientist to initiate an AI Challenge project with the emphasis on the deep learning.

What is the DeepReviewer? So, we all know there is a number of challenges the reviewer was facing. And the first, it's difficult to access the historical knowledge due to turn-over. And also, the reviewer really, the crunch on time, wanted to rapidly access to the relevant information from the public domain as well as from the internal resources. And lastly, and we need to have a way to maintain the institutional memory.

So, in order to address these issues, we decided to develop the AI framework to assist the review process. An essential the reviewer can quickly access the information they need for the review process. So, the way we see it when we develop such a system, we are really simulating how we learn. And the way we learn is that once we born, we go through several different stage of the learning and eventually we graduate from the university. And once we graduate from the university, we will be able to do a number of things. For example, we will be able to get into PubMed to retrieve the relevant information you

want, to do your work, and the text summarization. That means you read the entire document. You will be able to come up a few sentences to summarize what you read.

And questioning and answering. This is the one that we are doing every day in the Google. You type in where we are supposed to go to the vacations, and this is the answer going to be pop up. The last one called the sentiment analysis. And that means you read the document. You will be making a judgment of whether this is a positive towards one opinion or negative towards one opinion.

So, once you graduate, you are very much be able to do all of that, but you are not capable to do the review job. You need to have some sort of special training. So, what we decided to do is to develop a DeepReviewer. The first, we took a Google module. The Google module literally learn everything on the website, and we consider it as graduated from the university. And we feed them 100 toxicology journals with a little bit over around 280,000 articles, let that module learn. And it became the DeepReviewer.

So, we are testing this system at first on the safety assessment. And of course, we in the end of the day, we want this module to be able to do all of these

four. So right now, we only focused on the questioning and answering. This is a result I show here is a preliminary result and there's a lot of room for improvement, but it just shares some of the concept and the progress we are making this area now.

Now, on the left side is the Google module and the right side is on top of the Google module we learn from the toxicological journals. And you can see you just using the Google module, you're typing liver and it came up all of these organs. And so, what a Google learns, you want to know liver, they gave you a kidney or pancreas, and so on, so forth. But after you learn a significant amount of the toxicology journal, you're starting to see some of the terms we use in our area, such as in a hepatic, and the hepatocellular, and hepatocyte, and those things were starting to pop up.

And this is, again, if we put in acetaminophen and you came up all the NSAID drugs, but the DeepReviewer will be able to identify some of the term like APAP, acute liver failure, and the overdosing, and the related of acetaminophen. This is just a quick run using the commonly used tools. There is a number of the tools available. And not only just learned from the Google Document, you can also learn from the Twitter, and the Facebook, there are a

number of the tools are available, and we are systematically evaluating these tools. And for example, this is just a comparison between the Word2vec and the FastText methodology. So, as I said, we just started this project and we're still communicating with the Regulatory Centers as to whether they will find on the utility of these kind of the tools.

So now I'm going to talk about the machine learning. So, between 2005 and 2014 and during the MAQC Consortium activities and we collaborated with CDRH to launch a large project to evaluate the machine learning method for the genomics biomarker development. And there's a number of the paper published in Nature Biotechnology and the Genome Biology. I'm not going to talk about here. But I just want to point out that there is a couple things and we never really be able to address. And the first is about data size.

Now we know more samples always give you the better or robust models. The question is how many is enough? And the second, and we also know that adding more features usually on the courser issue called the curse of the dimensionality. But that the modern technology can take advantage of this. So, we never be able to address these issues. So, what we decided to do, we work with the

CAMDA. It's called the Critical Assessment of Massive Data Analysis. And this is a society to provide a platform to evaluate the big data analytics. So, we lead a CAMDA challenge project to evaluate AI and machine learning for predicting drug-induced liver injury using that genomics data. So, we have 11 teams from the nine countries which participate in this effort. And the general conclusion is that deep learning really outperformed the conventional approach.

However, we also realized the dataset we use is not large enough. So, during the pondering, what we're supposed to do and what's the next steps, the Office of the Chief Scientist at the Office of the Health Informatics, this is under the Office of Chief Scientist, come to us, they say, hey, so why are we just working together to utilize the larger dataset to evaluate the deep learning methodologies? And if you don't know about it, the PrecisionFDA has been around for several years now. And this is a tremendous part of forms established by FDA to allow a larger research community to working together to address a single issue.

And for example, on the Precision FDA already launched three different challenge. One is led by CFSAN, another by CDIH, and the last one by NCI. There are more

than 4,000 users to use this platform. So, we are starting to plan a project to assess the artificial intelligence and deep learning for the biomarker development.

Okay, so where we go from here? Several things that we wanted to do. And first, of course, continually develop the big data analytics and particularly in the area of the AI for the FDA data. And the FDA has a huge amount of documents. And I did not elaborate on some of the documents we have been using in our group, such as a lack of patient narratives, the meeting minutes, approve letters, and so on, so forth. And we are trying to develop various AI tools to mining this data to support the review process.

And the DARPA(?)labeling document is now a good examples and good dataset for the AI because that is over 100,000 in the draft labeling document. And going to be very suitable for the AI And we also wanted to study the computational reproducibility and we have been working on this project for a while now. And National Academy of Science just released over 300 pages of document to talk about the reproducibility. All they talk about is the computational reproducibility. And that means the same dataset that you use in different statistics will give you

the different results. Which one you going to trust? This has become a huge issue.

And we also starting to work with the electronic health records. And this is surely is the direction to go, particularly in the era of artificial intelligence. And this EHR data can be used as a real-world evidence to support the FDA review process. And we're going to continually evaluate alternative methodologies for predicting safety, such as DILI.

In terms of the support, and of course, are we going to do what we are doing right now and just ask a little bit more power on it, if we'll be able to hire the people. So, in the past few years we published a number of the papers. And most of these papers that's published is review papers and to summarize some of the ideas where we supposed to go in terms of the genomics technology or the rare cancer, a rare disease, and so on, so forth.

So, on the feedback requested, there is two urgent requests, really, from our Division or at least very urgent to our Division. First is how we can be able to recruit the talent and the scientists to our Division. And we, as I mentioned, at the very beginning we've been suffering for many years now and always running about 80 percent up to full capacity.

And we implemented several mechanisms. First of all, we focused on the local university. We bring the local university student to come to the NCTR, work with us. If we found that they are very good, we convinced them to work with us. And we also use social media such as LinkedIn. And sometimes we get a good hit. Most of the time is not. So now I just ask you whether you have some other recommendations.

And a second, working with electronic health records. This is extremely painful and expensive. And of course, you can for the reasons are very obvious because the deidentify issues. So, there are several on the publicly available electronic health records. And as for example, like MIMIC, those are the database that is publicly available. You need to sign a lot of the papers, but you can get that data as deidentified, but the scope is limited. And I just wanted to know whether you know any other database we can use. So, I'm just going to stop here. Thank you very much.

DR. LANZA: Thank you very much. We're open for some questions. Yes.

DR. RAMOS: I'm always immensely impressed by the directions that you take and the things that you do. So, I congratulate you on how this program has evolved over

time. And I think that what you're doing in the AI space will stand to make a huge difference as time goes on. I have one ignorant question and then one question relative to the data that you presented.

The ignorant question first. How are you differentiating between AI machine learning and deep learning?

DR. TONG: I do the shorter answer. So AI is a general concept about how we can train the machine to think and act like a human. You have a number of ways to train the machine. So, for example, you can use so-called supervised learning, unsupervised learning, reinforcement learning, transfer learning, those al; belong to the machine learning domain. But if you don't use all of this method, such as the one we use, it's much more like you look at a text document, how you convert it to some sort of the informations. It's not specifically and considering it's a machine learning and there are also several other areas. The definition is pretty blurred. Sometimes you do use the machinery learning.

DR. RAMOS: There is some overlap.

DR. TONG: Yes. Actually, AI is more like overarching concept and consider everything you are train the machine to do the jobs we do, they consider as AI Now,

deep learning, it's a very specific algorithm. And early days we call it artificial neural network. That means you have a three layer of the neuron; you have an input and output and in between is a hidden neuron that will do the other tricks. So-called deep learning just adding a little more layer of the neurons. And once you have more neurons to be added between input and output, you have many ways to manipulate how these neurons to be connected. So, data was the deep learning was come in play.

DR. RAMOS: More what we used to do when we were setting up networks of genetic interactions and so forth. The reason I asked you that question is because as you roll this out, you may want to add clarity to what you mean by the spheres that you're putting together. When I saw this slide and when I got the question, I Googled it and even Google didn't know the difference between the three, let alone the audiences that you're going to be targeting, and especially as you grow the program, your own desire to give it shape and definition for what you're doing. So that's a suggestion for you.

My question related to what you showed us, when you did the query comparison between Google and the tox journals, in some ways, I was not too surprised by the finding that you got because there is an inherent bias to

that comparison. And that is most toxicology is always done using an organ-based approach. And so, because of that, automatically you've cleaned your file, essentially, you've curated your data set. Whereas when you go to Google, anywhere that liver appears is going to populate. So, as you grow this and test the strength of your deep learning exercise, you may want to use a better comparison than Google. I would have been perhaps more impressed by the strength of the iteration had you done tox versus hepatology, for instance. Or tox versus something that's even more predictable than tox would have been. So that's just a little bit of food for thought for you.

DR. TONG: Fantastic suggestion. Definitely. And this is just meant to be illustrative which direction we are going. It's extremely preliminary at this point. But thank you very much for the comments.

DR.LANZA: Mary-Ellen.

DR. COSENZA: I hesitate to even ask this, but are you working at all or looking at what can be done with this SEND data that's been collected by CDER?

DR. TONG: Definitely is, yes. But we have not get our hands on the SENDs data yet. But fortunately, we are very closely working with the Office of the Computational Science, Lillian Rosario, she essentially

have a JANIS database was established to collect the SENDs data with the SAS transporter. So, in the future, yes, we will. Thank you.

DR. GANEY: Very nice presentation. I agree with Ken, that you're doing really great work. And I look forward to your DILIst. My question relates to that. As you know, a big problem in drug-induced liver injury is finding some clarity on which drugs do cause liver injury and what the severity of that liver injury is and where they fall. And you could look at all of those databases and there you can find a lot of inconsistencies or a lot of lack of agreement. So, is DILIst going to fix that? Or at least maybe attempt to address it? I'm just curious because I would really love that.

DR. TONG: I still remember 10 years ago when we started this project, try to put together a list of the drugs. And we will be able or confident to call which one cause liver injury if not. And we immediately realized this is a Pandora's box. You should never ask this question. So, we have (indiscernible names) on the phone, and after two hours, I still have no idea how to define how to determine which drug cause liver injury or not. And later on, John Senior came back, says you should look at the FDA drug labeling document.

Now, the labeling document itself has a lot of the problem and the way is how they put a labeling document together. But besides that, the guidance provided by labeling document, indeed try to capture the information provided to balance the view. So, after all this consideration, we decided using the labeling document as the standards to determine whether drugs cause liver injury or not. This is our first paper was published in the Drug Discovery Today, back in 2011. And of course, a lot of the comments and suggestions.

And one of the critical suggestions is, we did not do causality assessment. So, five years later, we did all the causality assessment which generate over 700 drugs. That means not only this drug cause liver injury, you have a causality assessment in place. Again, the causality itself is a debate because there is a number of ways to do the causality. So, and we finish that. And then when we come to the DILIst and we starting to look at the drugs and not including FDA drug labeling document and those drugs probably on the Japanese market, on the European market. So, we do not have a document to determine whether this drug cause live injury or not. So, what we did, we merged these drugs, published in the literature into our drug labeling based approach.

So, all in all, answer your question, everything we do is based on the drug labeling document verbiage with the causality assessment to determine whether is a liver injury or not. And some people's using so-called a case-based approach. That means number of the case has been reported for certain drugs. And some people's using severity-based damage. That means, how many people died to take these drugs. And some people are using enzymatic assessment, the transferase and ALT elevations when you reach a certain level. All these three methods have their own drawback. So, it is a little bit difficult. Yes. And I can just point to one thing, when we send this paper to review, usually get like a two-reviewer come back, we'll get a six. And the argument all of them argue the comments of what you just said.

DR. LANZA: Thank you very much. I think what I'll do is stop so that Dr. Heflich can go ahead and begin his program. We're right about 45 minutes from when we started. The last talk is the Division of Genetic and Molecular Toxicology and it's Dr. Heflich.

Agenda Item: Division of Genetic and Molecular Toxicology

DR. MANJANATHA: Good afternoon. I'm sure many of you must be surprised. This is not Dr. Heflich. I'm Manju Manjanatha, I'm Deputy Director of the Division of Genetic and Molecular Toxicology. And Bob Heflich, whom you heard this morning, is our sheriff. So, what I'm going to do is over the next 30 minutes give an overview of the Division research activities for 2019.

The staff. The most valuable asset of DGMT is our staff. And then compared to other Divisions, we have a relatively smaller Division. We have 27 FTEs or government employees or positions. And 11 of them are the permanent research scientists. And 10 of them support scientists. I guess I could make a statement that maybe we are the only Division where we have equal number of PIs and support scientists. And then three FDA staff fellows and one of them is externally supported and one FDA visiting scientist is also externally supported. And we have two admin personnel.

So, there's all together 27 FTEs. And then the rest of the members are authorized post docs. We have six a bomb and then three are ORISE Post Docs. We have six of them. And then three are externally supported. So, all together we have 33, which is one less of what we were last year. And some of these members appear on this slide.

All the external support for these Post Docs come from FDA/CDER, FDA/CTP, and also NTP.

As far as our outreach, we have the highest percentage of our collaboration with NCTR Divisions. I've listed these divisions here. Biochem Tox. In the parentheses are what sort of project or tasks that we are interacting with. With the DBT is mostly chemistry support. And we did some work with Mary Boudreau for carcinogenicity. And then lately, maybe Bob may have mentioned this, we recently got hold of Pac Bio II. It's an advanced next-generation sequencer which has more powerful technology than Illumina. And so, we are collaborating or sharing this with scientists from DBT as well as Microbiology.

We also interact with the DSB. That is the Division of System Biology, mostly to do with tissue models, both 3-D rat and human. As far as Division of Bioinformatics, mostly NGS data analysis. And we do interact with people from Microbiology, as I said earlier to use Pac Bio II instrument, and also Pathology mostly for the histopathology of 3-D animal tissues.

As far as the FDA Regulatory Centers, almost 30 percent of our collaboration with the CDER, CDRH, CTP, and CBER. And many of these Centers have presented their data

and then included some of ours in their study as well. As far as government agencies, almost 20 percent, we interact with NIEHS/NTP, mostly analyze system and also some botanicals that we are studying. And EPA, we don't have any current collaboration, I remember that one of our members represented as a gene tox expert at (indiscernible) meeting the EPA.

Universities, maybe 10 percent. The two listed here, the UAMS is University of Arkansas for Medical Sciences and University of Arkansas Little Rock. These are local universities; we interact with them. And the UMD is University of Maryland in Baltimore. Thanks to them because they maintain a Pig-A mutation database for us.

As far as our global leadership outreach, we have leadership roles in HESI, Health and Education Science Institute, International Workgroup for Genotoxicity Testing, OECD Committees, and also IAARC members is attending as an expert. And many of our members are involved actively in the SOT and the EMGS. SOT is the Society of Toxicology. EMGS, Environmental Mutagenesis and Genomic Society. And many of our members served as president, secretary, and then council all elected bodies. So, we are all heavily interacting or involved in these professional societies as well.

As far as our mission, our vision, it is to improve public health by providing FDA with the expertise, tools, and approaches necessary for comprehensive assessment of genetic risk. So, what are our goals? The most important goal is to respond to agency needs for chemical-specific data. Examples are like nanomaterials, impurities, and tobacco products. I guess this morning we have discussed some of these.

Second goal is to maintain DGMT's tradition of leadership in regulatory assay development and validation. Many of the assays listed here are developed in-house like Hprt, TGR, the transgenic mutation model. A couple of years back I developed one, a hairless albino model, for the purpose of using it for photo carcinogenicity studies. And then Pig-a, I don't have to say much about this because it's already been discussed, and we are doing serious effort into get Pig-A assay OECD test guideline acceptance. So, a lot of efforts going in this direction as well. And then the last goal is to establishes new paradigms for regulatory decision making that integrate measures of genetic risk with biomarkers of toxicity.

So, our research strategies are most importantly, we want to engage, or we have been engaging FDA Products Centers listed here: NIEHS, National

Toxicology Program, Health and Environmental Sciences Institute, HESI, and other national and international organization to set research priorities and develop better biological models for assessing human risk, using 3-D human in vitro systems and develop more comprehensive approaches for monitoring genetic variation using again this in-house developed assay, ACB-PCR, By Dr. Barbara Parsons' group, Next Generation Sequencing, and Digital Display PCR. These are all supposedly sensitive techniques to detect rare mutations. So, we have been using them.

And then lastly, develop better ways of evaluating data to determine human risk using such as dose response curves, benchmark doses, and point of departure. And we had some discussion this morning when Bob was responding to Subcommittee reviewer comments. So, we are although don't have a project with this intention, but we are collaborating with HESI and other groups who are using somewhat these benchmark doses and point of departure studies or data.

All right. I've been asked to list top three accomplishments. We had to follow the format that Donna suggested for the slides. For this year, I've chosen these three accomplishments and I'm going to read through it.

The first one is Barbara's group. They made tremendous progress on defining the mutational basis for the in vivo - oh, sorry. The first one is Vasily Dobrovolsky - for the in vivo erythrocyte Pig-a assay, evaluating mutation induction in bone marrow erythroids and granulocytes. And second accomplishment that I want to talk about is from Barbara Parson's Group. Demonstrated interindividual variation in cancer driver mutant fraction to identify mutations with the greatest carcinogenic impact in specific human tissues.

And then the last, but not least, is Tao Chen’s group screened genetic toxicity using metabolically competent human cells and high-throughput, high-content methodology using CometChip and MultiFlow technology. I'm going to expand on this a bit later, but we need to follow Donna's slide format, so I'm going to come back on this later.

So next three slides, I'm going to talk about really briefly, ongoing projects. This one is Xuefei Cao's ALI system. I guess it's been discussed in great detail. I don't want to get into a lot of details or technicality of this but suffice it to say that in addition to developing a panel of - this is relevant molecular and physiological endpoints for evaluating toxicity in organotypic tissue

models like this one. It's shown in this black ink. We also want to add some gene tox endpoints as shown here like the nucleus frequency comet to detecting DNA damage. I put the gene mutation in question mark, but I could add a corrected next-generation sequencing. But this is our dream, our big aim to set up or develop Pig-a assay endogenous mutation assay in 3-D models.

The second ongoing project that I want to spend some time on is by Dayton Petibone's group. And they are developing in vitro approaches for evaluating reproductive toxicity, including germ cell mutation. The slide shows here, on the left-hand side, is in vitro rat testicular organoid. And on the right is the in vivo testes. And these were labeled with the nuclei. And the blue colored organ is a nucleus labeled with the Zo-1 tight junction proteins. They are orange in color. So totally cell barrier are seen. But what is more exciting is that the formation of seminal vestibules in the in vitro system are organization that look pretty much similar to in vivo, which is exciting.

And also, Dayton's group, they have gone further to evaluate other types of cells. And they are excited to notice that spermatogonias, sperm cells, niche - it's not shown here though - were seen in vitro at the base of the

seminal vestibules. This is really interesting. So, we want to continue with this model and explore further.

The third ongoing project, I'm a bit excited about this because it's sort of a territory or a province where no DGMT has gone before. This Vasily Dobrovolsky's collaboration with UAMS, University of Arkansas Medical Sciences, with the oncology group where we are evaluating Pig-A mutant RBCs in 25 head and neck cancer patients. This is before, during, and after cisplatin-containing chemotherapy.

So, the goals of this project are to determine if rodent RBC Pig-a assay is predictive of the human response and increase. The second goal is to see if the increase in the Pig-a mutant frequency, whether it informs the outcome of the chemotherapy regimen, whether it's a success or failure. And the third goal is to, if there are changes in the Pig-a mutant frequency, are they associated with secondary malignancies in the long run. So far, the study involvement with cisplatin has shown that it is a potent mutagen in rat RBC Pig-a assay.

Continue with the data. So preliminary data from the collaboration showed the data in the table below. But so far what Vasily's group has done is collected blood from ten healthy donors. And the human MutaFlow Pig-a kit,

by Litron, is very sensitive. And also, you can do magnetic enrichment to evaluate Pig-a mutation in reticulocytes as well as total RBC. That was good news. But as far as the chemotherapy patient is concerned, we have so far only one cisplatin patient recruited for the study.

It's a 41-year-old hepatocellular carcinoma and tongue cancer patient, and he received cisplatin at 100 mg/MALE 2: on days 1, 22 and 43. Blood was tested three times and the data is shown here. I don't want to get too excited because we have just one sample, but it is also some issue with that.

So, as you can see, this is reticulocyte Pig-A mutant frequency. This is before the treatment. You can consider this as background control. And then the patient was treated with cisplatin, the mutant frequency started to go up. And what is unfortunate is, the day 65 data is important, because this is 20 days after the last treatment.

So, we wanted to see what happens to Pig-a assay, but unfortunately, the patient was no show. And upon talking to clinicians and oncologists, they suggested that this is the major problem. But I am not sure whether just here at UAMS or other places because after the

treatment chemotherapy, many patients don't come back to the clinic. So it is kind of hard to follow up and it is going to be not good for us because we want to collect data from all of these patients and hopefully they all will come back and we will have more data and then we can evaluate the goals that we have set up.

All right. So, the next three slides will show the details of the top three projects that I showed you previously. This is Vasily Dobrovolsky's study again. And they made an excellent progress on defining the mutational basis for the in vivo erythrocyte Pig-a assay evaluating mutation induction in bone marrow erythroids and granulocytes. So, what is the big deal about it? Actually, it is a big deal. It's a landmark publication because as discussed this morning by Bob and Susan as well, the OECD test guideline acceptance of Pig-A assay is extremely important. And we have been pushing in that direction.

And one of the member countries - there are 38 countries; anybody could ask anything. So, they told us that it is important for us to prove that the phenotypic mutants or the mutant phenotypes have real Pig-A mutations. Well, under ordinary conditions is very simple. You just collect the mutants, extract the DNA, amplify the gene, and sequence, and sure. But the RBCs are target

cells here. And as all of you know, red blood cells, when they reach the circulation system, they extrude a nucleus, so they don't have any DNA or nuclear material.

So Vasily, being as smart as him, he decided to reach out to the precursor cells. And as shown here on the right-hand side - I'm not going to go what all this lineage - but suffice it to show erythroid precursors and granulocytes were picked up, and of course they all have intact DNA. So, he exposed the rats to potent classical mutagens, like ENU, DMBA, and then collected these erythroid precursor granulocytes.

Did the Pig-a assay and sequenced the mutant and showed the types of mutations is very consistent with the test articles used. So, this proved the phenotype and genotype relationship. And this actually pushed our effort to get the OECD acceptance of Pig-A assay almost a mile. In fact, as discussed this morning, the OECD acceptance of test guidelines is a long and arduous process. And hopefully this helped move the needle a lot further.

This is a project that Barbara Parsons' group worked on. Essentially, they showed interindividual radiation in cancer driver mutant fraction, which can be used to identify mutations with the greatest carcinogenic impact in specific human tissues. So, as I indicated

earlier, Barbara's group has been using ACB-PCR. This is allele-specific competitive blocker-PCR. It's a sensitive assay developed in-house by them. And it can detect rare mutation, actually one mutation, ten to five, wild type. So, they use that technique or assay and come up with this summary. And let me drive attention to the graph on the right-hand side. So, they used the ACB-PCR and then they plotted on that X axis the variability or the standard deviation of mutant fraction of cancer driver mutations.

So, they selected two cancer genes, KRAS and PIC3CA, and two porons(phonetic) of each. And so, they plotted the standard deviation variability and they are all color coded. So, the breast is red, colon is blue, and so on and so forth. Yes. This is in the normal tissue. And then each data point, I guess, represent standard deviation of almost 10 to 20 samples. The cancer driver mutation fractions. So that the variability that you see, especially with the breast and to some extent colon, this is the normal tissue, is suggestive of clonal expansion of some of the cancer driver mutation in these samples.

So then, what they did? So, on the y axis, they plotted the mutant prevalence in tumors of the relevant tissue. This is the tumor hotspot mutations relevant to the tissue that are discussed here. And then what they

found there is a significant correlation between these two parameters. So that suggests that the carcinogenesis process is stochastic (phonetic) in nature. And so that same two tumors may not have the same type of mutation.

So based on these data, Barbara suggests that intraindividual variability can be used to characterize the impact of different driver mutations in different issues. And secondly, early clonal expansion of cancer driven mutation can be characterized using interindividual treatment group variability as a metric. And then lastly, a metric that combines many such measurements may be useful as an early biomarker of carcinogenic effect. This is nice work and it's been published as well.

The last top accomplishment project that I want to talk about is the work done by Tao Chen group. So, what they've done is they use this CometChip MultiFlow technology and used metabolically competent human cells such as hepaRG or primary human hepatocytes and evaluated genotoxicity of many scores of genotoxic and non-genotoxic carcinogens. So, these high throughput, high content technology is amazing that this CometChip has 96-well format. And then each well has around 400 to 500 micro-pores with a width of around 30 micrometer per well. And then you lay whichever cell that you want to test, HepaRG

cells loaded into these micro-pore chambers and do the Comet assay analysis. And the image shown here on the left-hand side is an ex post control (indiscernible words).

As you can see, the nuclei intact. There's not tail as seen here. And on the right-hand side is the cells that are exposed to a potent genotoxic chemical like MMS, which is the DNA damaging agent, and it induces comets or the tail showing the genotoxicity. This also she got published this year itself.

As far as our future projects, we want to develop and establish additional complementary rodent and human 3-D tissue models to bridge the data gap between the rodent and human responses for the test article exposure. This is extremely important because, as you know, that there are a lot of pressure on toxicologists to sort of move away from use of animals and embrace alternative systems. So, we need to bridge this quite a bit of data gap between the in vitro system and the in vivo. So, we want to work in that regard and continue in that direction and establish or adapt more genetic toxicology endpoints to complement the array of general toxicology endpoints developed for in vitro tissue models. I showed you one project on that.

So, some of the gene tox endpoints that we want to incorporate are Comet, micronucleus, error-corrected next-generation sequencing, as well as gene mutation for the ALI airway model as well as 3-D tissue models that we are working on. And then develop and in vitro approaches for evaluating reproductive toxicity, including germ cell mutation. I already showed a slide on that. And we want to continue with that.

And then use computational modeling approaches to use in vitro data to evaluate human responses. This is probably more relevant to ALI. The dose response in vitro to human relevance or human dose response for comparison, which is useful for CTP studies that the ALI is being used for.

Okay, the next two slides, I will talk about sort of futuristic studies that are just proposed, but although the second project, it turns out that it's not that futuristic. I'm going to talk about that. It's got approved and the work on that has already begun. Maybe I don't have to talk about that because it's not the future project anymore.

So, this is the work or proposal submitted to MCMi, the grant, by Dayton Petibone's group. Essentially, he's trying to develop a non-human primate testicular

model of Zika where there's sexual transmission. This is a busy slide. And then I may want to walk you through some of this.

The idea of this is essentially use this model of the Zika virus sexual transmission using microfluidic system so that the data generated will help evaluate the safety and efficacy of FDA-regulated antiviral products that have the potential to reduce or abolish the sexual spread of Zika virus.

So, I guess maybe Suzy mentioned about Tissue company that we are working with, collaborating with, as well as we bought the chip tech microfluidic system from them. And they are showing on the left-hand side on the top, these are tissue compartments one, two, and micropumps up here. And the bottom I think they're staying with the blue and the green to show circulatory system, microfluidic system.

So, you can load two tissues at a time and they're all through circulatory connected. And this is the pre-assembled ready to go system. And the C here is testicular organoid with Zika virus infected, and you can load in the compartment these testicular organoids. And speculation is that the virus particles shed from this chamber was joined through circulatory and reach the next-

door neighbor, the second compartment where you can load either non-human primate kidney spheroids or embryoid bodies.

And the advantage of this system is you can check the supernatant here for intermediate endpoints and also test the presence of Zika. And for a terminal endpoint, it can go into these bodies and evaluate for infection or the presence of Zika virus. So, the idea - this is all a schematic or speculation as to what might happen, if the project gets approved. I hope it will. Dayton and his group can start the work and then hopefully generate data that will be useful to CBER.

This is the one that I wanted to talk about as a future project, but since it got already approved and I think Carolyn from CBER, she talked about it, so I don't want to spend a lot of time. Essentially this is using ALI system that we are already using in DGMT. And information from Bordetella pertussis infection in the airway tissue culture probably will help CBER to understand the normal bacterial virulence factors and which can probably help improve existing vaccines.

So, these are all the ALI cells, ciliated cells and basal cells and goblet cells and hypothetical scheme

showing the Bordetella pertussis attachment through cilia and then entering into the ALI culture.

So, since this project is already approved and started, hopefully will generate some data which it will be useful for CBER.

I guess that that was my last slide. Thank you all for your patience. And if I could ask you as a feedback, what emerging sciences or technologies can you advise us to pursue and then what future directions do you recommend for DGMT that would impact the FDA? Thank you.

DR. LANZA: Thank you. That was excellent. Can we open it up to questions? No questions? Here's one.

DR. FELTER: This is maybe a comment/question that also connects with the talk that Goncalo gave earlier where I did not hear any update on where NCTR is with in vitro to in vivo extrapolation, PPK modeling, and how that might help inform doses that are chosen or concentrations for in vitro testing or for organoids in terms of being able to make some kind of connection between, say, a more traditional rodent study and now using organoid systems, how we're able to establish a connection between the doses. Are there projects ongoing in that area, either in your group or in collaboration with the DBT?

DR. SLIKKER: Thank you, Susan, for that question. And you're right, as of yet, we really haven't got into the modeling that's been done by Jeff Fisher and also Annie Lumen and several others that are working with them in a team way. But certainly, one of the questions there is, how to use modeling not only to set doses that could be useful for future studies, but also to extrapolate them between the in vitro setting and in vivo setting. And so tomorrow we may hear a bit more about that, but I'll say that there are some collaborative studies that are going on with other Centers and within NCTR that address those issues precisely.

You saw a little bit of the work going on with the arsenic, the pharmacokinetics studies that sort of established that there was a nonlinear dose response situation, which is very critical interpreting, especially the human data that's available out there. As you know, there are some human data available, but the exposure levels are very high and therefore this non-linearity comes into effect and has to be considered in the interpretation of that for any sort of safety assessment.

But also, this is being used to establish models that can be useful for developmental tox studies, especially where you're interested in fetal exposure and

you want to know about placental transfer. And certainly, Annie Lumen has developed some very interesting models there. So, I think there are several of these models that have been pushed forward and published. And tomorrow we may hear a little bit more about them. And if not, I can certainly fill you in at a later time.

But it is something that NCTR is invested heavily in, and we find it to be quite useful in a number of different levels. And we'll continue to develop that kind of modeling capability.

DR. LANZA: Yes.

(Off mic comment)

DR. MATTES: In terms of exposures in vivo versus in vitro exposures, I would say a rather boneheaded approach, which a lot of folks are using, including those in my group, are to look at CMAX levels, if you've got that information, you use that and then you do multiples of that and look at toxicity, in vitro. There are issues with that, admittedly, in terms of binding the plastic, in terms of going to high concentrations where you might see toxicity anyway. But suffice it to say, it's an ongoing problem.

DR. GAMBOA DA COSTA: So, expanding a little bit on how we have been changing the way we take on new

research programs. I mean, one of the crucial elements that we're starting to include more or less by default when we do bioassays is to include the elements of (indiscernible word) dosimetry, because otherwise you do not really know how to bridge the findings of the rodent bioassays into an individual situation.

An example of other approaches that we have been taking is what Bill was just alluding to, the work with cardiomyocytes. And I was involved in that work. It was being led at the time by Leping? I think it still is on the NCTR side. And what my laboratory did was looked into an array of, I believe, 10 or 12 different drugs. And we tried to ascertain which fraction was actually available for the cells. And it varies wildly. It was just not really predictable by the structure. I mean, some of the drugs were clearly lipophilic. And so, you expected them to bind to the plastics. But others, it was just really not predictable. So, we distinguish between the fraction that was really soluble, the fraction that was bound to the protein, and the fraction that had actually been lost, we can only presume.

But it's that sort of work that can enable you to bridge that from live animals into cell systems. So, the assumption that you just dump something on a well and

that it's going to be available for the cell, it's really a big assumption. But, yes, we're starting to think a lot about that. And to the point that I think that there should be a debate about whether a bioassay should be initiated in a given animal system without preexisting pharmacokinetics that they can locate you where you should be.

DR. LANZA: Further questions?

DR. WILSON: Am I allowed to ask a question? I may have missed it and I apologize if I did, but I wondered if you could explain sort of the rationale or the hypothesis for why you're developing this in vitro model for Zika virus, sexual transmission, when it's fairly well established that you can detect infectious virus in sperm and semen. So, I'm not sure what you're learning from this that isn't known. And again, I apologize if you said it and I missed it.

DR. MANJANATHA: I guess we have an MCMi representative. Maybe she'll be able to tell us why, if she can respond to this. Because what I say may not be the right answer.

DR. MCGILL: Hi, I'm Tracy McGill and I'm the director of MCMi, MCM Regulatory Science at FDA. I'm from OCET.

So, we had begun speaking with Dr. Petibone about the potential applications for the tissue system that they have. There is a general interest in these testicular chip models. For example, we have colleagues who are working with Ebola and we know that Ebola hides out in the reproductive system. And they are interested in really learning about sort of viral changes, evolution in that kind of immune-privileged tissue. And they're doing those types of studies in animal models. They're trying to learn what they can in the clinic, but they really would like to have some in vitro systems to do some of that work for all of the things that folks are talking about, the need for in vitro technologies to complement animal work.

And so similarly, as you know, we had funded a project with UC Davis looking at Zika virus distribution. And you're right, we do know that the virus hides out there, but along the same lines as the Ebola, this system would allow us to do some of that viral pressure evolution type of work in a microcosm. And again, in a way that wouldn't require us to do non-human primate studies, because, as you know, the primary models that they do use for Zika virus are non-human primates, just like the primary models that we use for Ebola are non-human primates.

So, again, really, I think looking at ways that we can get information in an in vitro system that speaks to one of the three Rs, basically reducing the number of animals maybe rather than ultimately the replacement, which is something we would all love to see. But it's going to be an evolutionary process to probably get there.

DR.LANZA: Thank you. Further questions? If not, then I want to thank everybody for their attention for this long day. There was a lot of great information and I'm going to adjourn this meeting. It will start again tomorrow, 8:00.

(Whereupon, the meeting adjourned.)