# National Center for Toxicological Research (NCTR)

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**P R O C E E D I N G S (9:00 a.m.)**

**Agenda Item: Call to Order and Welcome**

DR. GANEY: Good morning everyone. And welcome to day two of the NCTR Scientific Advisory Board Meeting. This morning we will be hearing from the remaining three division directors, followed by a brief break, and then we will reconvene for discussion. So our first talk today is the Division of Microbiology, and Dr. Foley you are on.

**Agenda Item: NCTR Division Directors: Overview of Research Activities**

**Division of Microbiology**

DR. FOLEY: Thank you again for the opportunity to discuss some of the efforts that we have had going on this past year as well as some of the things that we have got planned for this upcoming year within the Division of Microbiology. This is the same slide you probably saw about six or seven times yesterday about the disclaimer, that the presentation is mine, doesn't necessarily represent FDA.

So I look at our division, we've got a really talented staff within the division. We've got 25 FTEs. Right now that consists of 17 principle investigator type scientists. Four support scientists, although with the asterisk, you see we're in the process of trying to recruit and hire three additional, so we'll get that up to seven. And then four administrative folks, which includes myself.

And then with trainees, we have 10 ORISE post docs, graduate students, and the like within the division. So a total of 35 staff at the moment within the division. So like Bob said, we're one of the smaller divisions within the NCTR.

Even though we are a relatively small division, we do have a lot of expertise in the division. This wheel shows some of the major areas that we've got, the people who are in the division who are within these areas. So we have everything from host-microbiome interactions to some work in the environmental biotechnology area, nano, microbial detections, this would be looking at microorganisms and FDA regulated products, trying to develop more sensitive methods, more specific methods, and sometimes characterizing products that haven't been very well characterized in the past.

Virology, we've got, with the pandemic we ramped up some of the work with SARS-CoV-2, and we'll get to a bit of that later. And then antimicrobial resistance and food borne pathogens. And a lot of the work that we do in these two areas kind of cross the boundaries there. And we do have a lot of people with expertise in multiple areas, so you'll see names multiple times in here. And like I said maybe the people are doing antimicrobial resistance and

foodborne pathogens, or nanotechnology with microbiome, nanomaterials' impact on the microbiome.

So this is our division mission statement. It was expanded a bit recently. And so we do work, research, trying to evaluate the impact of a number of different compounds and stuff on the microbiome, again some of those developing methods for detection of microbial contaminants, the antimicrobial resistance and virulence mechanisms, I know that Dr. Tan talked a little bit about some of our collaborative work in that area with CVM.

And then some work looking at supporting areas like the Office of Women's Health, or the Center for Tobacco Products, and some of the nanotechnology initiatives within the agency. And a lot of this then is to help improve risk assessments for our product centers as they're evaluating different products that come or different scenarios that come in front of them for regulatory action.

So we tried to work to work better over the last number of years to try to support our division mission. And so one of the way we have done that is trying to enhance our research interactions with the different centers within the FDA to help understand or assist their research needs, and then, because we learn what their needs are, then trying to develop with their input research projects that

will help to meet the FDA's regulatory science mission. And part of that then is to help to contribute to guidelines and regulations within the FDA, and then we want to also strengthen our program management.

And so part of that again is trying to make sure that our research meets agency needs, looking at benchmarks, whether that's information that goes into guidance documents, peer reviewed publications, those types of stuff, that show the value that we are providing the rest of the agency, and making sure that we are doing that as well too.

And a key thing is trying to make sure our facilities and infrastructure is set to be successful in these areas, because our building is one of the older buildings on campus, has electrical limitations and those sorts of stuff, and trying to figure out the best ways to utilize what we've got to meet the mission.

We do have a lot of different outreach activities both on the global front with different working groups and different technical groups. We have people that are serving on a number of different editorial boards, contributing there, peer reviewing manuscripts for these journals, involved with guest worker programs. And that has gotten a little more difficult for international folks, but we continue to do what we can in these areas.

On the national side we're involved with a number of the different societies, like the American Society for Microbiology or the Society for Virology, and some of the more regional ones like the Arkansas Bioinformatics Consortium, and the Arkansas Association for Food Protection and the like there. And on a number of different government panels outside of FDA with NIH or USDA or some of these interagency workgroups.

And then we are really trying to be active too with a number of the local universities, trying to work with students and then also trying to develop that pipeline for talent, for postdocs and support scientists and those types of things. And we are trying to also expand that out beyond the state borders as well with universities and the region and within the country as well.

So when I look at our collaborations with the different centers, we have got a lot of our ongoing research projects involve different centers. I think we have got projects ongoing with every center right now except for Center for Tobacco Products, but if you look at the approved research concept kind of bubble there we've got one project that's in the approval process.

Dana talked a little bit about that in her presentation as well yesterday. And so we really try to make sure our research meets the needs of the agency in

working with collaborators and partners in the different centers there. And the number there is the number of protocols that have interactions with those different centers. And some of our projects have multiple centers on there. And so they are truly collaborative. We can sometimes be the hub for a lot of different research within the agency.

When we look at some of the division metrics, right now we have got 22 ongoing protocols, nine approved concepts that we're trying to develop protocols, and usually as those concepts get moved up we are hopefully having protocols rolling off that are being completed and technical reports or publications, those types of things.

From those we've got a couple of our approved concepts that have been put on hold because of other priority projects, and then we'll get those moved up as we have the capabilities to do that with personnel and the like.

We look at abstracts. For conferences last year we had 20 abstracts. At conferences this year we're going to have more than that. Probably I would say closer to 30 as people get more comfortable traveling after the pandemic. And then we had 29 manuscripts published, and so if you look at the trend that's one of the higher years we had over the last eight to ten years.

This is somewhat duplicative of a couple of the other slides already, but emphasis in the microbiome and the microbial contaminants, antimicrobial resistance. I know Dr. Patterson mentioned that as a priority in his presentation, again supporting OWH and the CTP and nanotechnology initiatives, and then helping with these risk assessments as well do FDA regulated products.

So I want to shift gears a little bit and talk about some of the specific projects that we've got ongoing, and at different stages as well too. Suzy Fitzpatrick talked about some of the tattoo work that we've been doing. And so this work has been going on probably for six or seven years within the division, where we have had these multiple different surveys of different tattoo inks, permanent makeup inks, and looking for microbial contamination.

And what you see from the pie chart on the right side there is we saw 35 percent of the products were contaminated. Even some of those where the bottle said sterile. So there is a real problem there when over a third of the products are contaminated with different microorganisms.

And so these are from a large swathe of different manufacturers, both domestic, and to a lesser extent foreign manufacturers. And these have led to a number of

different publications. I've highlighted two here on the slides.

Most of those other surveys were done with aerobic culture. And so one of our more recent focus has been on developing anerobic, you're looking at anerobic contaminants among tattoo inks. All of the work that I've been describing here is done by Seong-Jae Kim and Ohgew Kweon, they're the principal investigators on these projects.

So with the anaerobe work, again about a third of the products have been contaminated with bacterial growth. And the organism that was found most commonly was this organism Cutibacterium acnes, which is a potential opportunistic pathogen.

So the group there is in the process of working with CFSAN to develop a new project where they're looking at developing molecular based methods so you can have more rapid, more sensitive, more specific detection of the different microorganisms in these products. And again this is being developed with CFSAN, but we also have input from ORA because the goal of this is to move the molecular methods into the regulatory workflow that ORA has for tattoo inks.

We also have a multi-lab validation ongoing, which is led by Ashraf Khan looking at the non-tuberculosis

mycobacterium associated with tattoo related skin infections, and I think Sean Linder talked about that one as well too, in his presentation yesterday.

A new project, a relatively new project that we have got going on is looking at avian pathogenic E coli, the virulence of those organisms to identify targets for antivirulence drug assessments. This is being done with the Center for Veterinary Medicine.

The antivirulence drugs are a little bit different than say the traditional antimicrobial drug for treating disease. And so you're targeting the development of disease, not necessarily the infection, with the different organisms.

And so you may target things like the ability of the organism to attach or to invade the host cells, or produce toxins that lead to the different diseases there. The impact may be the uptake of different nutrients that prevent the organism from becoming a pathogen. And so maybe they'll wash out of the intestinal tract before they cause disease.

There is an interest in looking at these due to the big problem of antimicrobial resistance. One of the reasons we were asked to help do this project is that it was again a relatively novel approach, and this sort of serves as a prototype for CVM on trying to understand the

types of questions that they might need to ask drug sponsors as new antivirulence drug applications come in, to help with their guideline development.

So you may ask well why avian pathogenic e. coli for this project? Well it's a major problem in the poultry industry. It is also one where there is some fairly good data out there about different virulent factors and that sort of stuff. We also have expertise within the division with that particular illness.

And there was just recently a paper that came out, it was in the Washington Post, about urinary tract infections having potential foodborne origin. And there has been work with these APEC, which they get outside the intestinal tract, as a source of urinary tract infections as well too.

And so with this project one of the things that we've been doing, the initial step is to develop a database of different virulence factors from e. coli to see what are unique within these APEC that say commensal e coli that may be in the normal intestinal tract of humans don't have in there.

And so what we've done, and I don't have my pointer here, we've added to some of the work that we did with the salmonella virulence database that I talked about last year. And so we can then upload whole genome sequences

and get kind of an output like we show in the image here, where the green are the presence of the different virulence factors, and the white are those that are absent. And then we can see what are unique to the APEC so then we can target those with the antivirulence drug.

And then also a future step then would be to evaluate, all right, if we target those virulence mechanisms, are there unintended consequences like increased microbial resistance or virulence there that VBM would need to be looking for it in future applications that come in.

Another project that we've got ongoing with CVM, this is one that I'm the PI on, the APEC project, Dr. Jing Han is our principal investigator on that project. But this Plasmid Toolbox Initiative, what we are trying to do is look at the genes of plasmids.

And plasmids are outside of the bacterial chromosome, they often don't usually carry genes that are essential for bacterial function in there, but they do encode things like antimicrobial resistance or increased virulence. And so they have a major impact on public health.

But they also have a lot of genes that are not very well characterized on those. And so there has been a challenge because they don't have essential functions for

bacteria, that has been a challenge to knock out genes and that sort of stuff. And so we have been trying to develop what we call this plasma toolbox to develop approaches to both knockout genes on plasmids and cure plasmids as well.

Dr. Dereje Gudeta is a postdoc in our group, has developed a series of vectors like the one shown here that will allow us to knockout genes, for example the virB operon, shown in the bottom. And then we can go ahead and assess function with those genes taken out of the plasmid and try to understand the function.

The neat thing about the vectors that Dereje has developed is they allow for coulometric screening. So it's hard to see on there, but most of those colonies on the plate in the center of that vector ring are blue. The arrows are pointing to white colonies. The white colonies then would be those where the gene has been knocked out, so we can screen those to look at that.

And then along with knocking out individual genes we can also try to knock out whole plasmids as well. Plasmids are interesting, they have developed what we call these toxin/antitoxin systems on there, and so they encode for a long-acting antitoxins, so the bacteria lose the plasmid, which they can and do still survive because they don't contain essential genes typically in there.

But the plasmids have developed these TA systems. So they have these long acting toxins that will kill the bacteria because it sticks around after they lose the plasmid, and then they encode a short acting antitoxin.

And so because of that we try to develop this antitoxin plasmid, it has in the light blue there a series of antitoxin genes, so we can insert that in, try to cure, remove the plasmid that has the antimicrobial resistance genes on it, and then this is a temperature sensitive plasmid, and so we can alter the temperature and remove that, so you can then knock those plasmids out, and then we can assess the biological relevance of those plasmids.

And then we have the plasmids that we knock out individual genes that we can complement those back into the cured strain, and then easily assess their role in virulence as well too. So kind of a neat system we're trying to work on.

Dr. Huizhong Chen and Jinshan Jin are doing a number of projects on compounded drugs with CDER. One of the ones that they are working on is trying to develop ways to assess sporicidal compounds. And spores are a huge problem in the compounding industry because they are resistant to heat and desiccation, they tend to be around in the environment quite frequently.

So there is a need for sporicidal compounds. Well, some of them are better than others, and so there needs to be methods to understand how good the sporicidal effects of these are. So there are methods out there, but they have limitations.

Some of them utilize poor quality spores, and so you may have a false sense of security, that you've got poor spores, and you use a disinfectant or a sporicidal compound on it and it neutralizes those apparently, it may give you a false sense of security, or you may not have the right exposure times on that. So they worked to develop a test panel with an optimized spore preparation time and methods so that we can effectively look at sporicidal neutralization.

And then they've done work on this optimization, and right now they're in the step where they're trying to establish the standardized methods there for understanding the quality of disinfectants.

Another project, I talked about this last year, is some of the work that Dr. Sangeeta Khare and Kuppan Gokulan are doing on the NTP-funded project to look at developing a framework for the risk assessment of the xenobiotics and how they impact the microbiome and the gastrointestinal tract.

And so a lot of this work is being built on to existing studies that NTP or scientists within NCTR are conducting on different compounds. And so look at what are the impact of these compounds on the microbiome, or gastrointestinal permeability. It's providing a lot of good data on some of these compounds. And they have had a series of publications that are ongoing, and they are continuing these efforts as well too.

Just kind of briefly I'll go through a couple other projects. A new project that Dr. Kidon Sung is doing with the University of Arkansas at Little Rock is trying to evaluate how different nanoparticles can impact biofilms, or can serve as antimicrobial agents for different multi-drug resistant bacteria.

Also going to look at the cytotoxicity of these compounds as well too, because that has a potential for either if they have low cytotoxicity maybe more beneficial for use, if they have high cytotoxicity then that may be problematic as well for use in healthcare, food applications.

Dr. Kuppan Gokulan has been looking at evaluating nanocrystal drug formulations. He has been using primarily the drug Zileuton, an asthma drug where he is look at nano drug formulations versus the parental drug, kind of the standard non nano formulations of the drug. One of the

things that he has noted is that there are differences in the gene expression of a number of the different cytokines and those types of stuff, and there are also some sex differences as well too in the immune response, where a lot of times with females you see an increased response with a lot of the different cytokines.

Dr. Jinshan Jin again, with some of the compounding work, she has been looking at the compounded triamcinolone-moxifloxacin combination, which is often used for cataract surgery, and there have been a number of different adverse advents reported with that, and so she's trying to understand how the drugs interact with one another, and looking again at some of the toxicity issues, and then trying to understand is there a safe dose of the different ingredients in these compounded drugs.

And then I mentioned earlier some of the work with SARS CoV-2. Marli Azevedo and Bruce Erickson and Seongwon Nho are leading projects in this area where they're looking at trying to understand some of the cardiomyopathy that's been seen, and so they have developed a series of different cell lines that will either have intracellular exposure of the different compounds, and then looking at cytotoxicity, or having particles then where you can also have the extracellular exposure, and then looking at cytotoxicity, and have seen the expression of some of

the nonstructural proteins can lead to increased cytotoxicity.

Another project, I talked a little bit about the APEC virulence database. That was being built on the salmonella work. And so the Salmonella Virulence Gene Database has gotten to now we are beta testing the version of it that will be made publicly available.

And so investigators can upload whole genome sequences and then they'll get a readout of the presence and absence of different virulence genes. There's a way you can look at how similar the genes are, the different virulence genes to reference strains. This has been a neat tool. There has been a lot of interest from folks at USDA and other places in addition to FDA on some of these efforts.

We're working as part of an interagency group where some of these, the different virulence genes that we've identified our plan to go into the NCBI sequence analysis pipeline, so that as people upload sequences one of the readouts that they can get is these particular strains might have salmonella pathogenicity island one or two or five or those types of things to help understand potential virulence mechanism.

Also related to virulence, we've got a study where we're looking at trying to develop 3D cell culture

systems for both intestinal epithelial cells and macrophages to assess salmonella virulence. And the hope there is with this model that it will be a bit more representative of what we see in the real world. We're working on optimizing that at the moment.

So Dr. Mark Hart is continuing to do some of his work with the in vitro vaginal-tract model, trying to understand how lactobacillus can understand staph aureus, and some of the compounds that lactobacillus develop lead to limiting staph infections. And then on the bottom there we've got work that Youngbeom Ahn has been leading looking at better detection methods for Burkholderia cepacia complex.

And this was mentioned yesterday as well by I think Karen, where we're looking at trying to develop molecular methods that will be more sensitive, more rapid, that kind of stuff. BCC is a major issue because it can survive in water as well as antiseptics, and so a major problem. So that is molecular methods, some of them are based on LAMP or the Loop Mediated Isothermal Amplification, which tends to give more sensitivity than maybe some of the QPCR methods.

Some future projects. We've got again some more work with coronavirus, looking at some of the how complement and activation associated with that can lead to

coagulopathies. Dr. Feye is working on this, she took over for Dr. Wagner who retired. I guess the second one there I talked a little about earlier, that Dr. Gokulan has been doing, building on some of the zileuton work. Dr. Feye is also working on trying to develop a biosystem that will allow for understanding some of the impact of compounds on the microbiome as well to this immune dependent host cell free microbiome model.

And then a new project that we're in the planning process is again trying to develop and understand some of the plasmid factors on the dissemination of virulence at AMR.

So, some of our challenges. And I'm running out of time, so I'll go through these relatively quickly. One of those is trying to balance our ongoing efforts with emerging priorities, this with COVID. We shifted a lot of people to work on that important public health emergency, and that we do see where we have somewhat limited personnel and equipment and that kind of stuff, and so how do we best utilize that, that tends to be a challenge.

We've got some vacancies as well, too. Looking at our division structure, do we hire a deputy or branch chiefs to kind of back fill positions there. And then as we have people retire how do we best fill those and leave, our last go we had two people depart for another center,

another person retire, that were scientists. We're backfilling those with support scientists right now, and we have to look at going forward what is the best approach.

I talked a little bit about facilities, what do we do to best utilize the facilities that we've got, what equipment do we need for the future, those are challenges that we've got.

Computational biology, I think somebody had mentioned it was one to five bioinformaticians to bench scientists, and now it needs to be five to one. We see that as well too, where we need to have increased computational capabilities.

And one of the things that we're doing right now is setting up what we're calling a computational laboratory with workstations and those types of stuff and having software available to do that. We're also looking at training staff. There's the data forward initiatives, we've got some folks that have been involved with that to develop their bioinformatics skills.

Engaging centers, that tends to be a challenge, especially without having FDA in person activities like the Science Forum and those type of stuff where we have engagement like we had in the past.

And then again some of the balancing of research protocols of those that we initiate here versus those

requested from product centers. There sometimes can be a little bit of a friction there, where people want to focus on their interests versus some of the needs, and so we're working on that as well too.

So again, some feedback from the SAV potentially is are we meeting the needs of the agency, when we look at future directions for the Division what is the best way to transition into new areas, how do we best recruit folks to NCTR. And then looking at organizational structures as well too, is it better to have kind of one unit, or having branches. And so we have had some internal discussions on that as well.

The next slide, I think that's the last one. Thanks. And I'm happy to answer any questions, or any feedback that folks may have.

DR. GANEY: Thanks Steve. It looks like Chuck has his hand raised, and then Mary Ellen.

DR. KASPAR: Thanks. A couple of questions, so I can leave time for others. But with your work on APEC or the Avian Pathogenic E. coli, have you thought about doing any kind of genome comparisons between the Avian Pathogenic E. coli and the urinary tract infections caused by E. coli? And if I heard you correctly, if those strains are linked or similar and they're disseminated by foods I think that would be pretty significant. So any plans on doing that?

DR. FOLEY: Actually, that is one of the reasons why our initial work in this effort was to develop that database. And so what we can do then is go in, and we have already done this some with our CVM collaborators where we have taken strains from the APEC causing colibacillosis with some human strains from human infections. The initial one, they were just defined as human infections, they are isolated from humans, so they weren't delineated UPEC versus those.

But that is something that we want to do, because, and in fact probably ten years ago or so we put in a proposal for Office of Chief Scientist grant to actually look at that in there, and it didn't get funded at that point, but that has been a long interest to look at that to see, because there is a good potential my PhD advisor Lisa Nolan had looked at that a number of years ago as well too. There are a lot of similarities.

DR. KASPAR: One other question. On all your genome sequencing data that you're collecting, with salmonella and various other pathogens, is there thoughts or efforts at looking at defining pathotypes, or delineating these various strains into virulence groups?

And where I'm going is with the enterohemorrhagic E. coli, those have been broken down now into different lineages, and it appears that some of those lineages are

less virulent or avirulent. And I know there are similar cases with various serotypes of salmonella. So in looking forward, is that something that you're looking at when you're determining all these virulence pathotypes?

DR. FOLEY: Yes, that is a key part. We had a lot of interactions through Genomics for Food Safety, it's an interagency group that has USDA and us and NIH and CDC. That's a big issue, trying to understand virulence types with different salmonella. USDA is looking at the potential of regulating based on virulence rather than presence/absence of salmonella. There are certain serotypes like salmonella Kentucky that very common in chickens, causes relatively few human infections.

And we did a paper that was led by Center for Veterinary Medicine, and we used our database to analyze those. And there are distinct differences between the virulence factors between those Kentucky that are associated with human infections versus those that are predominant in poultry. So yes, that is an ideal thing that we want to do.

And so I was talking to Dr. Han, who is the lead on some of that work, just late last week about we were setup very well to do that with this database, and the hundreds of thousands of sequences that are available in GenBank that we could utilize, including lots of them from

partners within FDA, or USDA, that we've got pretty good connections with. So yes, that is something that we really want to drill down with as well. That is a key part.

DR. KASPAR: I would encourage you to keep working with USDA because if you could come up with such a system that would have a huge regulatory impact on the food industry.

DR. FOLEY: Definitely. We have had lots of good discussions with them. That's the way they are moving.

DR. COSENZA: Just a couple of quick questions. You talked about emerging issues. I am just curious whether your group was involved at all in a lot of publicity on the recent eyedrop contamination issue, or was that so basic your expertise wasn't needed?

DR. FOLEY: So we have not been directly involved with the eye drop thing. The compounding drugs, a lot of those are using ophthalmic, for ophthalmic use. We have been involved with that but not directly with the eye drop part. We would like to I think develop that capability with this rapid response thing, the rapid response type activities that Dr. Patterson mentioned yesterday, this would be something that could potentially fall into that if there is a need.

DR. COSENZA: That seems like it would be useful, considering that would seem to have gone on for quite a

while. My last question was you mention at the end you asked for feedback about the structure of your organization, but I am not clear because in the beginning you had that nice circle where it seemed like lots of people were working on lots of different projects. Are you organized by different groups, or is it one big group now?

DR. FOLEY: It has been one big group in there. Dr. Carl Cerniglia was the Director, I was the Deputy Director for several years, and then after his passing I was Acting. So right now they've dropped the Acting title. And now hopefully that position will be available.

And so we're looking at, there are a couple reasons to go in different directions. One, with branches it provides maybe a little bit, more interactions within the groups. But the flipside of that is where we have a lot of interaction already with different groups. So microbiome and food safety, there is lots of overlap there. And so it may be better just to keep one group.

And so we have had some discussions within the division. And there are some that are very happy with kind of the way it is, the majority I think are, and there are a few that prefer the branch structure. One of the things that we are going to do is have a strategic planning session, probably in May, to kind of have a little more deep dive in that, get input from everybody, and probably

talk to Dr. Patterson as well too about his thoughts, and kind of put a proposal together.

DR. GANEY: So actually this comes to one of my questions, and maybe you just answered it. So you had in one of your first slides, I think you entitled it strengthening research program management. And underneath that were several things like establishing metrics of success and others. So I am sure that your division has had those in the past. Are you just thinking now that you're taking over as the new director, it's time to take a fresh look at that and update it? What's behind that thinking?

DR. FOLEY: I think a little bit of it is, so one, a lot of the old metrics have been numbers of publications, and so not necessarily what is the public health impact per se other than a publication. And so one of the things I would like to do, some of it is probably some of my own interest in this, some of it I think there is interest from the other parts of the agency to better capture kind of the public health impact.

And so that would be, I guess looking at not necessarily a publication coming out, but is that data that goes for example like the APEC stuff, if it goes to CVM and they use that to develop a new guidance that goes out for people develop antivirulence stuff, there may not be a

publication on that in a peer reviewed literature, but that has a strong public health impact.

We in the past have done work with Center for Tobacco Products, where we provided a lot of technical data, and some of the papers haven't followed us as quickly because of some of the regulatory impacts, and so there are different kind of metrics like that, that are not necessarily easily captured with here is a peer reviewed publication that has got a DOI and that sort of stuff. So looking at those.

DR. GANEY: Any other questions for Steve? Thank you very much for your presentation. We will now move on to the Division of Neurotoxicology, and I see Dr. Talpos is getting ready.

**Agenda Item: Division of Neurotoxicology**

DR. TALPOS: Thank you. Good morning everyone and thank you for your attention today. My name is John Talpos, I'm the Director of the Division of Neurotoxicology here at the NCTR. In today's talk I am going to start off by talking about some strategic objectives of the division, and then talk about some recent research highlights, and end with a discussion of some new projects that we currently have in various stages of development. Of course the disclaimer. This presentation reflects my views and not necessarily those of the FDA.

So the Division is currently comprised of 28.5 FTEs. 13.5 of these are research scientists, staff fellows, and visiting scientists. So essentially our PIs in the Division. We currently have 11 support scientists. We have two administrative positions, including myself, and two ORISE post-docs at the moment. Now, you can see that we have multiple open positions on our org chart, and we are trying to aggressively fill these. We've had four new hires since the last SAB, and we're currently advertising for two additional positions.

We collaborate quite a lot with other governmental agencies of course, working with most NCTR divisions, and multiple product centers. We disproportionately collaborate with CDER and CFSAN. We are currently working with the National Toxicology Program, the National Institute of Perinatology in Mexico, the HESI Initiative on their Translational Biomarkers of Neurotoxicity Project, as well as the critical path institute.

When it comes to global leadership and outreach, we start locally, working with the University of Arkansas Medical Sciences. Several of our staff have adjunct positions there, and we do also collaborate with them. We also work with the University of Arkansas in Fayetteville, UTHSC in San Antonio, the Icahn School of Medicine in New

York, the University of Birmingham in the UK, and the Virginia-Maryland College of Vet Medicine, Blacksburg Virginia, as well as Augusta University.

The mission of the Division is to identify and quantify the neurotoxicity associated with FDA regulated products. Our goal is to provide the data and expertise necessary for crucial regulatory decisions. And to do this we use and develop translationally valid imaging approaches, alternative preclinical models, and cross-species metrics of brain function.

So when it comes to strategic priorities, really the emphasis in the division is on generating data that can be used to help support regulation. When it comes to our in vivo research, this is disproportionately regulatory-like developmental toxicity testing. And the reason for this emphasis on developmental neurotoxicity is because this is where the biggest data gap exists. We know a lot about toxicity in adults, but we have many more data gaps when it comes to adolescents and the perinatal period.

Now, with our vitro efforts, I do not necessarily expect this research to drive labeling anytime soon. Frankly I think the assays just generally aren’t there yet, in vitro, to do real regulatory-like testing. With that said, I think vitro work has the opportunity to add a great richness to our vivo work when it comes to projects

focusing on vulnerable groups as well as combinational studies.

Take for example opioids. A researcher in our group is about to start a very large vivo research project on the effects of opioids in the perinatal period. The problem is though that opioids are very rarely abused in isolation. If you're abusing opioids there's a good chance that you're also being exposed to nicotine, marijuana products, alcohol, and a host of other drugs. And in our in vivo studies we just don't capture this.

It is just extremely difficult to do these kind of combinational work in vivo studies. And I think this is where in vitro studies and in some instances alternative models can really shine, because this kind of work we can do in these paradigms, and in a way they allow us to get much closer to the clinical reality by looking at how these compounds are potentially interacting, and how this might cause a safety margin to shift.

We are also working on developing an implementation of new methods. So take for example regulatory endpoints. The gold standard for a neurotox study is an H&E stain. This is a technology that is over 100 years old and focuses disproportionately on assessing cell death. I think we can do better than this. So we're working on bringing on things like measures of synaptic

connectivity as well as T2 MRI based biomarkers in the Division, and some other endpoints. I am also trying to bring in functional measures for our in vitro studies, such as microelectrode array, calcium imaging, and synaptic activity.

Again, I think this is kind of like behavior in vivo studies if you will, and can also add a lot of information, so we don't need to just focus on cell viability, but rather we can start looking at cell functioning.

Finally, I am working towards adaptation of a minipig for neurotoxicity testing within the division. I want to get us to the point where we have MRI, neurophysiopathology, and cognition test battery, all established and running for use on the minipig.

For active projects we currently have around 40 in the Division. About 25 of these are experimental protocols, and we have another seven or eight there in other various stages of development at the moment. These at this point are basically all in collaboration with the other research centers, or other groups outside of the NCTR.

Our publication rate has remained pretty constant over the last five years. However, I don't think publications are a great metric for the division. And the

reason for this is I don't think they reflect agency impact, something that you heard Steve just talking about. So on this figure you have the top, the projects from FY2022 that used the most waiver hours.

I want to draw your attention to a couple of these, because I think these studies exemplify the point really well. These I believe are really well aligned with the Division priorities, with the Center priorities, and often highlight the unique contribution of the Division and NCTR to the FDA's mission. But what you'll also see is they're not resulting in a lot of anticipated publications.

If we focus on just the two cannabinoid projects, the two vivo cannabinoids projects we have here, they accounted for over 10 percent of our labor hours in FY22. And these are multi-year projects. So we're going to be billing numbers like this across multiple years. And we're only anticipating getting two publications out of all of this work.

I've seen preliminary data coming out of these projects. The data is nice, but it is also disproportionately negative results. Now, data of this type is very important and is very impactful, but usually null result data doesn't result in a lot of citations. So as long as we're doing work like this, I think our publication rate will remain low in the Division.

Within our PIs we do have a pretty broad area of interest, which does allow us to adapt to a number of different project types and needs as they come. And so you can see here on this pictograph of our current projects. Now at this point I want to transition to talking about a couple of scientific projects in more detail.

The first of these are two recently completed projects, one on the developmental effects of CBD, and the other on the acute neurotoxicity of a single dose of ketamine during adolescence.

So in this project led by Tim Flanigan within the Division, animals first started being dosed on gestational day six with CBD, and they were dosed all the way up until postnatal day 21, at time of weaning. We used doses as high as 350 milligrams per kilogram. However, at the highest doses we saw issues with litter viability. Because of this, we ended up focusing our analysis on doses only up to 250 milligrams per kilogram.

So we estimated that 100 milligram per kilogram dose results in serum values of CBD approximately similar to what you get with epidiolex use, whereas 15 milligrams per kilogram would give you an exposure that would be like a very high over the counter dose.

Now, Tim and co looked at a wide variety of behavioral endpoints, and this is a very large sample size

study. The first endpoints they looked at were shortly after birth, and they continued assessing animals well into adulthood, looking at tests of higher order cognition. They also used a large number of tissue-based measures.

Now, the titles here where you see asterisks is where some statistical significance was detected. However often there was not a clear dose-response relationship, and in other instances where significance was seen it was a complex interaction, it was difficult to figure out what was actually driving that significance.

So as a whole there was not a whole not of effects of CBD on doses that didn't affect litter viability. We hope to publish this data later on this year.

Moving to the acute neurotoxic effects of a single dose of ketamine. It is hard to believe now it has been 10 or even 15 years ago when a series of studies came out showing that ketamine was extremely effective at providing acute relief for extreme depression. This was really seen as a breakthrough in psychopharmacology.

Because of the profound impact that ketamine was able to have in this patient group, there has been a desire to use it for other indications, and in a younger patient population. The problem is that ketamine has never been approved for use in children. It is very commonly used in

them as a form of anesthesia, but it never was approved for this use. So there is a lack of safety data.

Because of this lack of safety data, the Agency has been hesitant to allow clinical trials at higher doses of ketamine. It has been limited to less than what can be used in adults. So we set out to address this data gap, to see if there was any evidence of altered sensitivity in young animals to the effects of ketamine.

So to do this we treated animals at postnatal day 21, 30, or 35. This was our adolescent cohort. To a single dose of ketamine. So this would, depending on which model you used, this would be the equivalent of say a four- to 12-year-old human, when it comes to brain development. We also had a 90-day group, which was our adult control group.

Now, what we saw was that we only found necrosis in the PND 90 females, even though our positive control was effective at all groups. If you look at the figure on the right, this is our Cmax value, so we actually had the highest levels of ketamine exposure in our youngest animals, suggesting this wasn't an effect on bioavailability, that the younger animals were actually less sensitive to the effects of ketamine.

What was also surprising is when we look at norketamine levels, we saw that they were much, much higher in terms of terminal half-life in area under the curve in

the PND90 females, the only group where we saw toxic effects, suggesting it might actually be norketamine, the main metabolite, and not the parent compound, that is driving neurotoxicity.

So, moving on to ongoing projects, I want to talk a bit about our work with the T2 MRI as a biomarker for nonclinical neurotoxicity safety studies, as well as the vitro study looking at the developmental consequences of early-life exposure to opioids and cannabinoids that I alluded to earlier.

Serguei Liachenko has done a great job over the last 10 years putting together a dataset describing how T2 MRI can potentially be used as a biomarker of neurotoxicity. Neuro, be it tox or science, is a tough space to work in, because we can't actually measure the brain directly at work like we can with the heart. We don't have the equivalent of cardiac output.

We have to extrapolate from other measures to what the brain is doing. So if we want to go and measure neurotoxicity the gold standard is to go and remove the brain from the animal for that analysis. This means that we can only take one assessment at a time ever from one of our animals.

And the problem with this is on top of this many markers of neurotoxicity are ephemeral, you can't always

detect the effects of an insult days later. So what Serguei is advocating to do to get around this problem is to, prior to planning a classical histopathology study, is to perform MRI on a smaller cohort of animals, but to do repeated assessments.

So take for example the case of hexachlorophene, which is what you can see here on this figure. Serguei imaged animals repeatedly, day three, day six, 13, and 20. And what you can see is that on day six the brain lit up like a Christmas tree with these animals. But if you had done your assessment on day three or on day 20 it is possible that you would have gone and missed this effect. So what he is advocating for is not to replace histopathology, but to use this approach to augment it.

So in this instance you would go and perform your MRI and see that day six is your most vulnerable period, so you would then focus your histology on day six, and you would also know the regions of interest based off of your MRI analysis. Hopefully this can make your neurohistopath safety study faster, use less animals, and potentially be less expensive.

So I presented this project last year at the time Serguei still had a substantial to-do list as you can see here on the right. In the last year he has made quite a great deal of progress.

The area that I am most excited about with this progress has been on data collected for week neurotoxicants, or neurotoxicants at low doses. So here we are trying to see if the threshold of MRI ends up being similar to the threshold for a histopathological study. And if so this will really I think show just the power of this approach. At the same time, Serguei is actively preparing his letter of intent for submission of the biomarker, and hopefully that will get sent off this summer.

Next, I want to talk about some work that Dr. Fang Liu in the Division is doing with the effects of methadone and buprenorphine in combination with cannabinoids on human neural stem cells. So as you all know we are still in the midst of an opioid pandemic. This has been made only worse by the recent COVID pandemic.

And opioid use are going up, including unfortunately in pregnant women. At the same time we know that pregnant women are also using cannabinoids. And this number is likely only to increase with the mood to decriminalize cannabinoids.

And we also see that in fact some women are actually using cannabinoids to treat opioid withdrawal during pregnancy as they try to decrease their overall opioid consumption.

Dr. Liu's main question is whether treatment for Neonatal Opioid Withdrawal Symptom, also known as NOWS, can be improved. So to do this she wants to determine the effects of opioids or cannabinoids on human fetal neural stem cells and cells differentiated from neural stem cells.

She also wants to know what is the impact of using cannabinoids to treat opioid withdrawal. So she hopes to determine the effects of coadministration of opioids with cannabinoids. And endpoints include cell viability, receptor expression, as well as developmental cell fating.

You heard Fred talk a bit yesterday about this issue with metabolites with CBD. And this is something else that I'm hoping Fang will be able to resolve. So a primary metabolite of CBD is seven carboxy CBD, which reaches much higher levels in humans than in rodents. In rodents in contrast we see higher levels in seven hydroxy CBD. And this leads a great deal of uncertainty when it comes to our safety assessments.

Now, in a perfect world we would just take some seven hydroxy and seven carboxy and do a study with just those in rodents. The problem is these compounds are extremely difficult to synthesize, and they're very expensive to purchase. Realistically, a study, a vivo study of this type, is just impossible, because the compounds are too expensive. However, we can test them in a vitro system.

So what we have here is some very recent data that Fang was kind enough to share with me for this presentation, where she is comparing the effects of THC, CBD, and its two major metabolites in a pair of viability assays.

What I want to draw your attention to is these two datapoints, with the carboy and the hydroxy versions of CBD. Because what we can see is potentially a three to tenfold shift in potency between these two major metabolites, highlighting the importance of getting the modeling of these metabolites correct for neuro work in an in vitro setting, as well as vivo for that matter.

While this work is still ongoing it appears that these compounds are disproportionately affecting the growth phase of the cell cycle, not division. Now this slide is quite complicated, but the take home is that these effects appear to be additive. If you take two of the metabolites at a sub threshold level, put them together, you are then able to see an overall effect on viability. So it looks like these compounds are all having additive effects.

And I think when we move to vitro work, at least in the neuro space, this is an important consideration, because you need to think about the total exposure load, and not just that of the parent compound or the primary metabolites when assessing neurotoxicity.

Often here at NCTR, or at least in my division, we put together large in vivo projects with direct regulatory endpoints that are just too expensive for us to start based off of Division funds alone. We have to find a source of external to NCTR funding, be that one of our granting bodies or funding from one of the other centers to start this work. So we have had two projects on the shelves that look like are finally getting off in FY ’23 and FY ’24.

The first of these is a project led by Andrew Chen, A Comparison of the Developmental Effects of Different Treatments for NOWS. So this is funded by the Perinatal Health Center of Excellence in FY23, and it is in collaboration with CDER. It compares the therapeutic and toxic effects of morphine, methadone, and buprenorphine in rat pups that have previously been exposed to opioids. So we have this if you will addicted component, and so we can actually look at the effects in animals that more accurately reflect the clinical situation when it comes to treating NOWS.

This work will include PK to confirm dose levels, and Andrew plans on using cross fostering to reduce the impact of the maternal influence on these studies. And this will be a very large project. In total he is looking at

using 320 rats to study the immunohistopathology and behavioral effects of these various opioids.

We have another project looking at the effects of acetaminophen. Acetaminophen has a reputation for being safe during pregnancy. The reality is this has never been demonstrated, it is used by half of all pregnant women, and a number of epidemiological studies have come out questioning its safety.

This is a project that has been included in the CDER budget for FY24. I am hoping it will get funded, although it is always possible the budgets will change between now and the next financial year. We want to investigate the effects of a therapeutic exposure to acetaminophen in guinea pigs during the third trimester of brain development.

We plan to bring breeding in house, and this will also include PK. This is a large study where we will have 192 prenatal guinea pigs. And I am quite excited about this project because it represents a first here for us at the NCTR working with the guinea pigs to perform a tox assessment.

So moving to entirely new projects, I want to talk to you a little bit about some planned work that Josna Kanungo wants to do with the developmental effects of heavy metal on the zebrafish, a blue sky project led by Serguei

Levchenko on assessing Olney lesions with MRI, and another project looking at validation of microelectode array technology by Syed Imam.

So started with the electrode array project, this is the one that has progressed the furthest in our internal protocol tracking system. For those of you who aren't aware of MEA technology, it is a pretty cool idea. We have these very tiny electrodes, and we can then grow cells in culture on top of these.

And this approach allows us to measure the tiny electrical pulses that are released by neurons. Through this approach we can look at neuronal functionality by looking at the mean firing rate, excitability by looking at burst of action potential, as well as connectivity by looking at synchronous firing.

Now, Syed is setting himself up for long-term success by focusing on a validation dataset. So he will be working with a lot of classical compounds that are known to influence neuronal functioning, but that we don't necessarily expect to be neurotoxic. So this way we can get an idea of the dynamic range of these assays pharmacologically, and then this will really help us when it comes to assessing toxicants that don't result in an acute cell death.

Syed will also be looking at culture conditions. What we often forget about as neuroscientists is that many amino acids are also coagonists of NMDA receptors, and so you would expect that these amino acid concentrations should have a direct impact on the performance of your culture, and potentially on the toxic potential of whatever your manipulation might happen to be.

The ultimate goal of this project, one of them actually is just to stay current with MEA technology. This is an area that is moving very quickly. And it does seem that MEA is getting some traction as a regulatory endpoint for seizure liability. Not sure if that will happen, but it is definitely an area that we're interested in watching. Internally, we want to use MEA to study seizure risk, and I also hope to see this incorporated into many of our in vitro studies to assess synaptic function and overall neuro function.

This moves me to Josna's project in heavy metals in zebrafish. This is an area that Josna has been working in for the last couple of years now. Suzy alluded to some of this work yesterday. She has done some fantastic work looking at arsenic in isolation, and one of the things that she has discovered is that arsenic looks to be having some of its effects via the sonic hedgehog pathway.

One of the components that I really like about Josna's most recent studies is that in conjunction with Fred Beland's group, she was able to get some if you will internal dosimetry out of the zebrafish. They went and measured overall levels of heavy metals in this arsenic work within the fish.

And even though Josna was working with a dose that was many times above the EPA level for safe drinking water, these aren't necessarily causing radical increases in internal heavy metal concentration. So at her lowest dose where she saw changes on neuro endpoints, we're talking about somewhere between a 10 and 20-fold increase in arsenic levels. So again, high but certainly not out of the realm of clinical relevance. So her most sensitive endpoint happened to be a reduction in motor neurons.

And Josna is working on using this finding to develop an OECD adverse outcome pathway, describing how arsenic can cause changes in sonic hedgehog expression, how this will impact motor neuron development, and how this could then eventually be relevant for developmental disorders such as ASD.

Now, what Josna wants to start next is investigating the impact of cadmium in conjunction with arsenic. So the first question is does cadmium impact the endpoints that she previously saw with arsenic. If it does,

does it also go and intersect the arsenic AOP? Again, we will be working with Fred's group to determine internal exposures, and also a little bit of a blue-sky element to this project, we were working with the Divisions of Systems Biology to see if regional levels of heavy metals can be measured with MALDI imaging. This is a little bit of a longshot because we would be doing this in zebrafish embryos at 72 hours old, and these are really very small animals, so this might not be feasible, but it would add a whole new level of confidence if this is something that we could do.

The next project I want to talk about is assessing Olney lesions with MRI. And before I can do that I need to talk a little about MRI in the minipig. And it is important to remember that when it comes to MRI, bigger is better. And that is because MRI resolution is a function of scan time and signal to noise ratio, or SNR.

Now, this image here is comparing the pig brain to that of the rhesus monkey to that of the human. And what you can see is the pig brain really is pretty big. It is approximately the same size as the rhesus monkey. The data I am about to walk you through was generated in past studies with the rhesus monkey, but I am hoping these values will hold true for the domestic pig, and they will

demonstrate how using the pig offers a distinct advantage for this particular study.

So what contributes to SNR? Well, voxel volume, size of the receiver coil, and magnetic field strength. In the minipig we can have a much larger voxel. Based off of the nonhuman primate, we're talking about 16 times bigger. This is a real boon for our signal gain.

Also unfortunately though we will need a bigger receiver coil, which was going to result in approximately 5X loss in signal. And we will also be forced to use our 4.7 Tesla MRI as opposed to our 7.05 Tesla, which will mean we will also have a loss in field strength resulting in about a 1.5 loss in overall signal strength.

Combined though this does mean that we end up with double the resolution in the nonhuman primate, and hopefully also in the minipig. The minipig is a species we are going to perform this work in. When compared to the rat.

This then gives us two options, where we can either have a faster scan time, or we can have a smaller slice. And for this work we'll definitely go for the smaller slice.

The reason that we want the small slice is that Olney lesions are very small, we don't know if we can actually do this. So Olney lesions were first described in

the late 1980s, they develop after treatment with NMDA antagonists, and you see that vacuoles develop in certain cortical regions hours after exposure. Now, these vacuoles are actually within the neuron. So I'm not expecting that we can detect individual neuronal changes, no, nothing like that, but hopefully that we can detect a fingerprint associated with these changes.

Now, these vacuoles seem to disappear a couple of hours, or the next day, after exposure to NMDA antagonists. But they appear in regions where you frequently see necrotic neurons, say three days post exposure. Which leaves this question do Olney lesions precede cell death. It has always been a real difficulty knowing exactly what an Olney lesion means from a regulatory perspective.

DR. GANEY: Hi John, you have two minutes left.

DR. TALPOS: Well our plan is to expose animals, to perform MRI at three hours, and then basically look to see if we have a fingerprint, and then figure out those animals that have Olney lesions, and then go back and see if there is a fingerprint. We'll then go and follow this up at 72 hours. And basically, the idea is to see if we can use three-hour MRI to predict seven hours histology.

So challenges for the Division is determining which alternative models and new technologies to invest in, maximizing the regulatory value of our in vitro research,

and maintaining the deep expertise and a flexible workforce, which is what we need to do to be adapting overall to changes in in vitro work.

Areas of specific feedback, the one area for me really right now is should we be working to actively develop adverse outcome pathways. I never really thought of this as deliverable in its own right, but when we think about combinational work like what we're seeing with the heavy metals, I don't know how we address this issue without maybe focusing more on adverse outcome pathways and less on specific tox deliverables, but I am very happy to hear thoughts about that. And of course, thank you to the Division, this isn't possible without all of you. And our many fine collaborators who help us in our work who are not on this slide. And that is everything, and I apologize for my length.

DR. GANEY: Thank you very much for that presentation. Mary-Ellen, you turned on your video. Does that mean you have a question, or a comment?

DR. COSENZA: I have a comment on the adverse outcome pathways. We had a session at SOT that was sponsored by IOTOX, the global collaboration coffee, and it was actually sort of a topic of discussion, this whole question of endpoints versus adverse outcome pathways.

I don't really have an answer. Most people actually were in agreement that they sort of worked together, but it was a really interesting discussion on the pathway. So I applaud you for looking at that. I don't really have an answer to the question, but it definitely seems worth the effort. But it does seem to take years to actually get that done.

DR. GANEY: Ken, do you have a question?

DR. RAMOS: Great presentation, John. I have a couple of questions or comments for you. I appreciate your comments about the use of MRI as a way of maybe improving upon pathology findings.

But a word of caution I guess for you to consider is that focusing the pathology assessment strictly on when the areas that you see MRI signal picking up might actually lead you to wrong conclusions, because you do not yet know exactly sort of what the physiological, biological implications of that signal is in the MRI, and you could actually be missing important windows of pathological response by not doing it.

I do appreciate the idea that by using the MRI signal as a guide you actually can bracket pathological analysis in a way that can actually then be informative to what is a transient lesion or a potentially transient

lesion, or what is a long-term impact. So more to come I guess as you guys engage in that particular set of studies.

DR. TALPOS: For sure. I am interested to hear what kind of feedback we are going to get when we let this out into the real world if you will, because we have talked a lot about it internally, but that's our kind of groupthink, and we are going to be appreciating input we get from everywhere once this expands a little bit further.

DR. RAMOS: Yes. The real challenge here is you really don't yet know how to interpret the MRI signal and what it actually means in the context of pathology. So that's going to be, I think the interesting part of the exercise that you're going to be engaging in.

And then my second point relates to the ketamine findings that you were looking at. I presume that your interest in the Olney lesions stems from some of the ketamine set of studies, since a lot of the NMDA is going to be connected to that. But my question is have Olney lesions been described in humans.

DR. TALPOS: No. So, this is an interesting point, and I skipped over this. They have never actually, as far as I know, been described in a large primate, in a large animal in general. So that's one of the reasons we want to do this, to see if it actually even shows up in the minipig. It has never been mentioned in the monkey as far

as I can tell, nor has it been, there have been some weird reports of stuff that will happen in extreme abuse type situations, but as far as I know it has never been shown anything even close to a normal dose in humans.

DR. RAMOS: So, when you think about translatability of those findings, how are you expecting to be able to achieve that?

DR. TALPOS: That is a good question. Part of the issue right now is is this just a theoretical concern or is this a real concern. I mean, to begin with, I think if we don't see them in the minipig with whatever compound we work with first, we're going to have to follow that up with the second or third to see if really it looks like it's not translating into a higher order species, maybe somehow this is a rodent specific phenomenon.

And then afterwards it is just whether we can start to say yes, if it does occur do these neurons go on to die. It may turn out that the finding is not translatable at all. I think that would be fantastic if that is the case.

DR. RAMOS: I hear you, and I appreciate your idea that going to a higher order brain might actually be the way to begin to assess that. Of course, it is going to be important to make sure that when you communicate the findings you do so in a way that makes it clear about

translatability, because otherwise you could be raising a can of worms that is unnecessary.

My last point, if I could, I was intrigued by your calculation of labor hours. I appreciate very much, the intent. I think it's very important, so I support your approach. How did you do that, how did you actually track those hours?

DR. TALPOS: This is a little dirty, it just came out of our time recording system. So every two weeks we have to go and say how many hours we worked and which projects we worked on. So it is imprecise, but it comes out of NCTR just labor recording systems.

DR. RAMOS: So essentially self-reporting.

DR. TALPOS: Yes.

DR. RAMOS: I suppose that as you guys continue to explore ways to measure impact, this process is going to be somewhat refined.

DR. TALPOS: I would suspect so.

DR. RAMOS: Because otherwise it is going to be not a reliable resource for assessment.

DR. TALPOS: You get some folks who never put any Division overhead for instance on there, and you know that's not realistic, they're not constantly working on projects, so there certainly are cases over over-reporting. And other times people might put something as Division

overhead because it's well I'm working on this piece of microscope and maintaining it, but that ends up being extremely important for Project X.

DR. RAMOS: Thank you so much.

DR. GANEY: Luis, do you have your hand raised?

DR. VALERIO: I am with the Center for Tobacco Products, and just want to get your thoughts on potential test strategies for seizure liability assessments, in terms of in vitro or high throughput perhaps integrated with computational methods, to see if you have explored that. The thinking is to assess aerosol type mixtures or ingredients.

DR. TALPOS: We have not thought about doing aerosolized compounds yet. I think that is something that could be done. I am always a little bit wary about extrapolation from that, because at the end of the day that is not how these compounds reach the brain.

By the time they get to the brain they're in the blood. So I'm not sure, if that's the only way you can do the exposure physically, I guess that makes sense. But I would think if possible, doing the liquid-based exposure is probably better than doing aerosolized.

Going forward, we would be very happy to work with you guys on seizure liability. This is a new model for us, but if you notice some of the compounds that we have on

there, there is a lot around acetylcholine receptors, and I know you have a lot of other concerns besides just those. When it comes to the computational side that is something I haven't thought about at this point, although as the assay gets up and running, I suppose that is a bridge we would eventually have to cross.

DR. VALERIO: Thank you. I appreciate it.

DR. GANEY: I have one comment on AOPs for you, John, which is there are many ways that chemicals can affect normal physiology. Some of them result in toxicity, some of them lead to adaptation. But although there are many ways that can happen, those ways are not infinite.

And so when you start trying to piece together like you mentioned, trying to merge two AOPs to see if you could get a picture, I think that that becomes a bit murky, especially when you throw in the adaptation effect. So I would just, if you're really going to go down that pathway I would caution you to keep that in mind as you're moving forward.

DR. TALPOS: That point is very well taken. And AOPs are relatively new for us. When it comes to this heavy metal question, I don't know how we do it without AOPs. That's not to say that the AOP is the holy grail and the solution here. I'm just kind of stuck scientifically figuring out how else to answer that question.

And it seems to me like the best solution. But it could be Josna is going to go and work on this project and a year from now I'll just say no, no, we were crazy, we've got to find something else. But thank you for that input, it's true. It's a new area to me, I don't know much about AOPs at this point in time.

DR. GANEY: I guess we are ready to move on to our next speaker. Thank you very much for your presentation. And we will now hear from Dr. Schnackenberg about the Division of Systems Biology.

**Agenda Item: Division of System Biology**

DR. SCHNACKENBERG: Good morning. I guess I get to finish up the division talks before we move onto the subcommittee later this afternoon. I am Dr. Laura Schnackenberg, I am the Division Director for the Division of Systems Biology. I have been in this position for about six months now.

As some of you may know that have been on this committee for a while, Dr. Mattes was the Division Director from about 2014 until the end of 2021. And then last year we did kind of a rotation through division directors. So Dr. Rick Beger actually gave our division overview last year.

So just the disclaimer, which you've seen before. So we can move on to the next slide please. So this is just

a list of our current division staffing. And I do want to note that this does not capture our vacancies in terms of our FTEs.

And we also have a number of open spots for ORISE, postdoctoral fellows that we are trying to recruit. I think we have about five of those. We did have one staff member leave last year, primarily for family reasons and needing to relocate. We had one retirement. Dr. Desai, actually two retirements, but Dr. Varsha Desai who many of you have heard from in the past retired at the end of the year along with Carrie Morland who worked a lot with Varsha.

So currently we have 17 research scientists, staff fellows, and visiting scientists. Nine support scientists. Four administrative positions. And one commission core. We do have two ORISE postdocs currently, for a total of 33 within the division.

And so this just shows our division organization. And so this is current as of the last year. We had already kind of consolidated from three branches into two as of the last SAB last year. But in our immediate office I am the Director, and then Dr. Jessica Hawes is our Deputy Director.

We have two branches, we have the Omics, Models, Imaging, and Chemistry Branch, which was previously known

as the Biomarker and Alternative Models Branch. And this was led by Dr. Rick Beger. And then we have the Innovative Sciences and Technology Branch, which currently I am still serving as the acting branch chief there, and working on filling that position.

So this just shows some of our outreach. And what I really want to illustrate here is that our division has been very proactive in the last several years, making it a priority to establish and foster collaborations with our product centers.

And this kind of goes back to one of the questions that was asked yesterday I believe of CDER, is how do we prioritize these. And there are some cases where we will develop kind of a protocol, quad chart, and kind of go out to the other centers and say hey, this is what we're thinking about.

But we are trying to move more towards a method where we are engaging those product centers ahead of time before we even develop that quad chart and try to figure out what their needs really are, and with our expertise how can we help them out.

So this is just some numbers from our last year. We did have 48 active protocols in 2022. 17 of those did include collaborations from the other NCTR divisions. And you will see that the majority of those projects, 46 of 48,

included collaborators from our other FDA regulatory centers and offices, including CDER, CBER, CVM, CDRH, and ORA. And you did get to hear about some of these efforts from our other centers yesterday as well.

We do collaborate with other government agencies, including NIH and NCATS. We have a number of academic outreach efforts as well. Starting here as John said with the University of Arkansas for Medical Sciences, we have an ongoing collaboration with the Medical College of Wisconsin, the University of North Carolina Chapel Hill. We have a number of projects ongoing with the University of Tennessee Health Science Center doing a lot of our COVID work because they do have BSL3 facilities there. The Medical University of South Carolina, Arkansas State, and Georgia Tech.

We also have a number of global leadership outreach efforts. And this just lists a few, I am sure we are missing some here. A big one is the involvement of Dr. Rick Beger and Li-Rong Yu with the metabolomics, quality assurance, and quality control consortium.

We have folks working on a number of different HESI working groups, including the Stem Cell Working Group, Development and Reproductive Toxicology committees, the Genome Working Group. We also have people working on the HEIS U01 Advisory Team. And then we also have somebody on

the Organization for Economic Cooperation and Development, Expert Group on Developmental Neurotoxicity.

And so this is just some of our select collaborations. Some of these you will hear about later or in the subcommittee review. But just kind of to show just a little bit of flavor of what we're doing, we do have pandemic related research, that is with collaborations with CDER and CBER. One is looking at perinatal, therapeutic, and vaccine nonclinical studies with SARS-CoV-2 variants of concern. We are also looking at differentiation of immune symptom response. You heard a little bit about this yesterday from Dr. Rick Beger's projects.

And in collaboration with CDER, CBER, and CDRH, we are getting ready to start a multi-center biomarker qualification study of anthracycline associated cardiotoxicity. With CDER, CDRH, and CVM, we are developing a predictive toxicology folliculogenesis model.

With CBER, you heard a little bit yesterday, we are working on looking at the safety and efficacy of CAR-T cell therapy. With CVM and CDER we have a project that will be looking at cannabinoid neuropharmacology and pharmacokinetics.

And finally, with CDER we have a couple of different efforts looking at microphysiological liver systems, one to evaluate adaptation and regeneration after

a toxic insult, and another one to evaluate multiple liver MPS platforms. And as I said, many of these will be presented in more detail for those that are participating in the subcommittee meeting later.

So what is our Division mission and goal? Our Division mission is to address regulatory research needs, knowledge gaps, and emerging health threats using systems biology approaches and innovative technologies in areas of interest that I'll talk about here, but this is certainly not limited.

But these areas may include safety and use of medical products, drugs, biologics, and devices, safety of foods and supplements, safety in detection of components and impurities in regulated products, and the development of technological standards and methods used in regulatory science.

With the goal being to use these cutting-edge approaches to address knowledge gaps and regulatory or safety concerns of interest to the FDA product centers and offices.

And some of the strategies that we are using include a number of state-of-the-art tools to characterize these systems biology and therapy induced toxicity or disease. We run the gamut of omics technologies, from

transcriptomics, epigenomics, metabolomics, proteomics, lipidomics, and imaging.

We are also developing spatial proteomics and a pipeline for that, and DBB has been very helpful in getting that up and going. We're looking at new alternative models, both human and animal based. We are using in vivo disease and pharmacodynamic models and other pharmacological tools. We are also incorporating innovative computational and instrumental technologies, with the goal being here to evaluate differences in risk in toxicology related to species, tissue, sex, and subpopulations.

So these are just some of the metrics from the previous year. As I already mentioned we had 48 active protocols. We also have 7 active support protocols. And theses support are also used to kind of support the needs of the other centers as they come. And this is kind of along those lines, some of these is that rapid response tool that Tucket alluded to yesterday. So just being able to quickly respond without having to go through the full protocol.

We had 20 scientific publications,24 oral presentations, eight of those were internal to FDA and 16 external. 29 poster presentations. We did have 11 competitive intramural funding awards, seven CDER central funding awards, and four COVID supplemental funding awards.

Our staff overall received 17 accomplishment awards in the past year, and then we did participate in 18 FDA working groups and subcommittees, and then 20 external working groups as well.

So now I would like to go into some of the examples of our ongoing projects. One of them is the effects of SARS-CoV-2 infection on pregnancy and prenatal and postnatal development. And I will talk a little bit more about that in a bit.

In vitro assessment of opioids on neural precursor development and proliferation. Opioid induced defects on lipid distributions. Putative protein biomarker predictive of anthracycline-induced cardiotoxicity. The development of this mouse model of inflammation to evaluate CAR-T therapy. And then the evaluation of leishmania parasites as potential vaccine candidates.

So the first one I want to talk about is the COVID-19 effects. So the aim here is to identify and understand the potential adverse effects of COVID-19 during the perinatal periods. And so this may provide hazard risk assessment data for SARS-CoV-2 infection that can lead to enhanced safety for pregnant women and pediatrics.

So we know that with SARS-CoV-2 infection there are still a number of questions that need to be answered: what are the effects during pregnancy on both the mother

and the fetus, what are the effects on fetal organ digenesis, what are effects of maternal infection on term infants, and are there effects on the adolescent sexual maturation after exposure in utero.

Additionally, for those mothers that are treated with remdesivir, what are the effects there, does the remdesivir alleviate those potential effects, or do we see no change. And obviously a lot of these questions in humans and children that were born of mothers that were infected with COVID, this is going to take a lot of time. So we are trying to do this in animals and see if we can translate that to children ultimately.

So our nonclinical perinatal SARS-CoV-2 infection has a risk assessment that includes looking at pregnancy and maternal health, vertical transmission, embryo/fetal development, postnatal development, juvenile development and maturation, and then the long-term effects. To this end we're looking at several different endpoints, including clinical pathology and histopathology, we're assessing developmental endpoints, and then doing functional observational battery.

For this we are using a mouse model of the human ACE2, where the human ACE2 cDNA replaces the endogenous mouse ACE2 sequence. And so we've got this ACE2 knock-in pregnant mouse model. This will allow endogenous mouse

regulator elements to directly express the human cDNA. We did select to use the delta variant for SARS-CoV-2. At the time this study was being developed that was the primary variant of concern, and we do know that that variant did cause severe impacts on pregnant women.

So in the case of delta infection, pregnancy was associated with an increased whisk of ICU admission, and for increased risk for invasive ventilation compared to nonpregnant women. There was also increase risk of death observed for pregnant women as compared to those nonpregnant.

And so we did a couple pilot studies before we started the pregnancy study in order to determine the correct inoculation dose, and also a small pilot study within the pregnant animals to make sure that we did have the correct inoculation dose chosen.

So our treatment groups, and this is an ongoing study, include our control group that is not inoculated with SARS-CoV-2, we have the 25,000 plaque forming unit group, and then that same group also treated with remdesivir. And so you can see the study design over on the bottom left slide. And so we are infecting on gestation day seven, at which point we're getting baseline weight and temperature. And then treating with remdesivir for one to ten days after infection.

And we're also collecting samples at both GD11 and GD18. And so our endpoints here do include that body weight and temperature from post implantation, day one to eleven. We are collecting organs, including the lung, heart, spleen, brain, kidneys, uterus, placenta, and umbilical cord. And then we are doing pregnancy assessment, which includes gravid uterine weights, number of resportions, number of life fetuses, and number of dead fetuses.

So in this study as I've said it is ongoing, we have completed one batch, with the second group getting ready to start very soon. But what I want to point out is that the clinical symptoms that we've observed thus far have been observed, but they're comparable to controls. We don't see much change in body weight across the various groups.

Important here what I want to point out in this table is that we do see a greater post-implantation loss in the two inoculated groups. And we do see that maybe in the remdesivir group there is less post implantation loss compared to the group that was inoculated and not treated. Again, this is ongoing, this is just a smaller group of animals, so we'll see if this holds throughout the study as we move on.

So moving on to one of our studies of opioids. In this case we're doing an in vitro study to look at the effects of opioids on neural precursors. So the aim here was to determine whether direct exposure of human induced pluripotent stem cells to opioids influences neural differentiation, proliferation, and viability.

And what I want to point out on this slide, initially four cell lines were looked at to determine the different effects, and we looked at IPSCs on day zero, the effects on embryoid body formation from day zero to five, neural rosette formation from days five to 12, and then following that neural precursor cell expansion from days 12 to 19.

And you can see the various opioids listed on the right that we tried in the study. And the bottom panel just shows some of the typical measurements that were done, gene expression and NPC proliferation are the two most common. We don't always use the microelectrode array to assess neural function, but we did in some cases with this study as well to see the effects there.

So in this slide what I really want to point out is if you look at the lines with the triangles those represent the NPC proliferation. And what we see there is that the opioids did not really seem to infect the proliferation of the neuro precursor cells. However,

several of the opioids, codeine, methadone, and morphine, did have a greater effect on the IPSC proliferation, and the NPC differentiation. So those were the two most impacted.

So this just shows some of the effects on the neuro rosette formation. And over on the left side you see the two panels, we have control, and we did also treat with valproic acid, which is known to be teratogenic, and toxic.

So what we see here is that methadone and codeine did cause issues with the formation of the neural rosettes, whereas the other three opioids in this case did not seem to cause much impact on the formation of the neural rosettes. And this was done at you can see very high doses, 300 times the Cmax and 1000 times Cmax.

And this just shows some of the gene expression changes just with one specific opioid. This was methadone at 1000 times Cmax. And you can see over on the right the major biological processes that were impacted during the Rosette formation, including gland morphogenesis, axon guidance, neuron projection guidance, CNS neuron differentiation, and others.

We did also have an in vivo arm of this opioid exposure study. And so in this case what we did here is we used our Matrix Assisted Laser Desorption Ionization

Imaging Mass Spectrometry to look at some of the fetuses after opioid exposure.

And so the aim here of the in vivo study was to determine whether that maternal opioid exposure increased the risk of neural tube defects. Which may have helped address the data gap, including approaching if certain opioids pose a greater risk to the developing fetus, and identify the most susceptible neuronal cell types, which obviously we're also doing with the in vitro study.

So in this case we had our fetus which was sectioned at about 12 microns thick, we applied a matrix, and then did imaging on that. We also did have a section that was stained so that we could correlate the lipid changes with the H&E staining.

And so we just see two different examples here. So we have a methadone, a fetus collected from maternal exposure to methadone, and one after exposure to morphine. And what you see is that they both cause dose dependent distribution changes in normal brains for lipids linked to hypoxia. And so hypoxia is one of the proposed mechanisms by which toxicity occurs after maternal exposure. So in this case we see that PC, phosphocholine 34:2 was increased after exposure, while phosphocholine 34:1 was decreased.

We did observe exencephaly in some of these fetuses, but we did not show the data here. And it was very

difficult to compare both those fetuses with the normal brains and the exencephaly just due to the different sizes and developmental differences between those. And we are still working on some data analysis and additional analyses in these studies.

Another study that I want to talk about is validating these putative plasma biomarkers of anthracycline induced cardiac function. And so the aim here is to identify, verify, and develop new clinical plasma biomarkers to predict cardiotoxicity.

So as many of you know, anthracyclines, and especially doxorubicin are commonly used for the treatment of breast cancer, but the cumulative dose can result in cardiac damage in women down the road. Oftentimes this is well after treatment has ceased. So, can we identify some biomarkers that can be predictive of those who ultimately are at a higher risk for cardiac damage?

And so initially this was done with a smaller sample set. This was done in 83 patients, which was split into a discovery set and a validation set. We had 39 in the discovery set and 44 in the validation set. And these patients had undergone four cycles of doxorubicin plus cyclophosphamide treatment. Blood was drawn before treatment, and after cycles one and two.

A left ventricular ejection fraction was also measured in these patients before treatment and after cycle four. And then they were grouped by normal and abnormal LVEF validation. Aptamer based proteomics were used for discovery, and Olink proteomics for analytical validation.

And so you see kind of the study design at the top at the right panel on the slide. And in this initial study seven potential protein biomarkers were identified that were significantly changed in both the discovery and validation set. And so we are now moving into this larger validation study which will include multiple centers throughout the United States that we'll receive samples from.

Another area that we're working on is the CAR-T cell therapy. And then our part here is to assess the acute, inflammatory toxicities following CAR-T administration, whether it be ultimately cytokine release syndrome, which is well known, neuroinflammation, or neurotoxicity. So as many of you know, CAR-T cell therapy, genetically modifies a patient's T cells to express those proteins that specifically recognize and kill the cancer.

And so just kind of a little description of how this is done shown in the bottom right panel where you collect the blood,

isolate, and genetically modify that T cell, expand, and then administer the CAR-T cell therapy to the patient, with the idea that hopefully these cells will attack the tumor cells and kill the cancer.

So we have successfully developed a mouse model here at NCTR to look at the effects of the CAR-T cell therapies. Our approach here was to identify soluble factors mediating the inflammatory responses and develop those nonclinical models of CAR-T cell therapy associated inflammation and toxicity. And then ultimately to conduct toxicology and safety assessments for potential CAR-T cell therapies.

So the initial studies really were just to develop that model, make sure we had the appropriate tumor model in the mouse, what was the appropriate timeframe in order to inject those CAR T cells, and then follow up and look at the inflammation and neurotoxicity markers.

I apologize for the missing legend on the graph in the middle. The circles are for those groups that did not receive CAR-T cell therapy. And you can see that we did see a sharp decrease in the body weight with the tumors. And then the triangles are from those that received CAR-T cell therapy. And so we see that body weight holds fairly steady up to 80 hours after CAR-T treatment.

Also on the right we see the un-transduced group, and then we see the serum IL6 level at 24- and 48-hours post CAR-T cell treatment. And we do see that after 48 hours there is a much larger increase in the serum IL6 compared to the other two groups.

And I think this is the last one I want to talk about. But this is the evaluation of potential leishmania vaccine candidates. And so the aim here is to evaluate the safety and immunogenicity characteristics of novel leishmania parasitic vaccines. And this was listed as one of the 20 neglected tropical diseases by the WHO in 2019.

And so in the bottom right panel you see a heat map of metabolite changes in the neutrophils. And so we have our control group, and then we have our leishmania centrin group. And in this case the centrin was deleted, which the centrin gene is responsible for the enhanced pathogenicity. It prevents the parasites from replicating in the animal.

We are hoping that by deleting that gene you're not going to have the response that you would. And so you see your control and also your wildtype, and we see very similar changes in the metabolites in the neutrophils for those two, with a very distinct pattern for that LmCentrin group in the middle.

This is another study. This is the effect of light on plasma metabolites. And Dr. Sun will go into this in much more detail tomorrow, and you heard a little bit about this yesterday. But this is the effects of light treatment on both platelets and plasma. And the right side should actually be platelets there, not blood plasma. This is to determine whether this is an effective method of preventing bacterial growth within those components.

And so cluster one is representative of antioxidants, photosensitizer, metabolites, and cluster two includes some vitamins and other steroid metabolites. I won't get into too many specifics here, but the results do indicate that with this light treatment that platelet aggregation is not affected, and that the membrane activity remains intact. And again, for those of you this afternoon that will be on, you will hear much more about this.

So before I wrap up with some questions, these are just a few of the future projects that we have upcoming. We're looking at a systems biology approach to delineate mechanisms of opioid addiction, evaluation of the perinatal cannabinoid central nervous system activities and toxicokinetics, developing a human cell based immuno-cardio new alternative model. Again, as I mentioned that multicenter biomarker qualification study.

We are also working with CDER to look at the assessment of hepatotoxic potential oligonucleotide drug impurities. We're developing a model to predict adverse events using drug endogenous ligand target networks. And then finally looking at evaluating multiple liver microphysiological platforms and how they work with different liver cell types for drug metabolism and hepatotoxicity studies.

So you've heard about a lot of the challenges, and our challenges are no different than any of the other divisions or NCTR as a whole. But a major one is how do we balance emerging research needs with our ongoing research. I think we all did a great job of pivoting with COVID19, but at the same time we did have ongoing research, and how do we balance those and make sure that we're not dropping projects.

A major challenge within our division is obviously maintenance contracts. So we do have a lot of high-end technologies, a lot of mass spectrometers that carry a heavy price with them and do tend to need service quite often, and certainly that yearly maintenance. But how do we balance supporting our protocols with also making sure our instruments are up and running and not pouring all the money into those.

Communication obviously with the product centers. Like I said I do think we're getting better, and really trying to engage. But still it's a challenge finding the right people to talk to at some of the various product centers, and how do we overcome that.

So just some of the feedback. And I think this is probably fairly similar to what you've heard from many of the other divisions. But one of the big questions obviously is are we effectively addressing the needs of the product centers, what approaches in addition to our current efforts might better inform us of the FDA research needs at the other centers and offices.

Are there other emerging sciences and technologies that you advise the Division to pursue? We do have a lot of efforts in the omics obviously in developing new alternative models. We're primarily focused on liver and cardiotoxicity models, as well as those developmental and reproductive models. We've done a lot of work in those, which many of you will hear about later. But are there other models that are of interest, kidney, other type of things that we may be missing, are there other future directions that would be recommended for our division to move forward in?

And I think that's it. So I think I'm about on time, maybe I have a couple minutes. But before I turn it

over I would just like to thank everybody for being here today, the members of the science advisory board, the representatives of our FDA centers and offices, Dr. Patterson and Dr. Mendrick, Dr. Hawes, my Deputy Director, Dr. Rick Beger who is our OMIC Branch Chief and has done a great job with that over the years, and then obviously the Systems Biology staff, we couldn't accomplish any of this without them. And my contact information should you need it. So I will turn it over to you, and I am happy to answer any questions.

DR. GANEY: Thank you Laura for a really nice presentation. Are there any questions from SAB members for Laura?

DR. MENDRICK: Laura, this is Donna. I just want to note that Tucker is now the Director, not the Acting Director.

DR. SCHNACKENBERG: You are right. The problem was Donna, I had to send these slides to you before that change got made. So I apologize. I am sorry about that.

DR. GANEY: I guess if we don't have any specific questions for Laura, and she is going to get grilled later today anyway, we can move on. Thank you very much. So what we will do now SAB members is we will take a 15-minute break and reconvene at a quarter after the hour, whatever

your hour is. And then we will have a discussion of NCTR research. See you in 15.

(Break)

**Agenda Item: Discussion of NCTR Research**

DR. GANEY: Okay, welcome back everyone. This is the time during which the SAB members have a discussion with NCTR about what we've heard over the last day and a half. Ken, you look like you're unmuted. Do you want to start?

DR. RAMOS: I was just making sure I was not muted before. So this still is open session, right? So we are just basically providing general observations relative to what we have heard?

DR. GANEY: Yes, that is true, it is.

DR. RAMOS: I will go ahead and get started. I actually sensed an improvement in this particular meeting relative to previous meetings. I have been trying to identify why I have reacted that way, and I don't really know for certain sort of what that impression, where it's coming from.

But I sense maybe the best observation that I can make is I sense an elevation in the level of energy across the organization that's not consistent across all of the groups, but certainly in many instances. I sensed a renewed sense of energy. I presume that this might actually be

related to having new leadership and being reinvigorated by that. Maybe it's related to FDA related organization changes that have reenergized people, but I sensed that, and I hope that actually is true.

The second observation is I continue to be disappointed by the unevenness in sort of what I perceive to be the quality of the programs across the different divisions. I think it is a problem that we have wrestled with for many years, certainly all of the time that I have been on the board that certainly has been the case. And I think some of the suggestions that we have made over the years trying to add some evenness across the presentations has helped, but I think more remains to be done.

An area where I think some attention needs to be paid to is this whole issue of impact. If you noticed, I am sure many of you noticed that it was a repeated theme across the different division presentations. But the interpretation of the impact varies across those divisions. And so some consistency in how impact is defined in my opinion would be helpful.

The idea that publications are not a good metric or a good indicator of impact, I receive that idea with mixed emotions. The reason I say that is because I totally embrace the idea that a paper by itself could be totally

meaningless if it's not something that translates into action in some form.

But by the same token, publications and peer review of the science that has been conducted within the center is something that is needed, and is I think something that is part of the metrics that have to be utilized to track progress of the division, for nothing else I think the depth of review that's associated with peer review is required to validate the quality of the work that's being done.

So that plus whatever else is utilized for impact I think is going to be an important area in my opinion for future attention by the leadership of the center. And I don't mean this just in terms of Tucker, I mean it across the division directors and everybody else.

I was intrigued by John's use of labor hours. And then of course all the nuances that go along with labor hours. But I think that that is an area where some attention I think would be important.

And then the last thing that I will say is that I hope that in future presentations one or two slides be devoted to innovation. I see the NCTR as a hub for innovation for the FDA, that certainly has been the case in many instances, where a lot of technology and approaches

originating from the NCTR permeate and drive the agenda for the FDA, for the agency as a whole.

And so if the divisions work to devote one or two slides to innovation, I think that will not only help them focus energies in areas that can actually be transformative and impactful, to the point of impact that we were just talking about, but also help guide our ability as a Scientific Advisory board to weigh in and maybe help support and enable some of those aspirations and investments that are made by the center. And I will stop there, any maybe circle back if there is time after we finish up.

DR. GANEY: Okay, Tucker, I see you have your hand raised. If you have something brief to say that's great, but I feel like we should get to the rest of the Scientific Advisory Board.

DR. PATTERSON: Certainly. I just want to mention, because I'm sure this will be on the mind of the other board members, but putting metrics to research impact is certainly difficult, and the agency has struggled with that, so much so that in the last two years I have sat in on a research impact working group with representatives, with probably some of these that are representing the other product centers today and yesterday on the call.

And so trying to standardize an approach when from a research perspective of course yes we want a peer reviewed manuscript, that's kind of what we desire out of that. But as I think it was Dr. Foley pointed out, sometimes work that the agency needs, the work that we are doing is being driven by a product center, that may not be the ultimate outcome. And so we're trying to standardize metrics.

Again I think as Michael pointed out yesterday we are trying to even define stakeholders. Who are the stakeholders in this? For us a lot of times the stakeholders are the other product centers. For the product centers the stakeholder is the American public.

So we're looking at what those outcomes are, what those drivers are, and we are trying to really standardize that. CTP has done an excellent job of really kind of standardizing who their stakeholders are, what the metrics are, and so the rest of us are struggling with that right now, but we are working on that for sure, Ken.

DR. RAMOS: Thank you, Tucker.

DR. GANEY: Thank you. I am going to go around my screen. Mary Ellen, you're at my top left, so you're on.

DR. COSENZA: Like Ken, I too was impressed with the level of not just enthusiasm, but also seemed more coordination between the centers and the divisions this

time than in the past, which is something that having been on this board a couple years now we've highlighted in the past, that we wanted to see more focus on that. And I think that certainly came across today.

Which I think is actually even a little more impressive considering what we've gone through with the pandemic and the lack of sort of in person interactions. So the fact that that seems more coordinated I think is actually pretty impressive in light of the physical constraints. I think that's the main point I wanted to make on the overall coordination for now. We will obviously have a lot more to say when we get into the subcommittee work later.

DR. WALKER: As a new member of the committee I was mostly trying to listen and learn. I think what has resonated to me were Ken's comments. And just to elaborate on that, I think we all realize impact is a little in the eye of the beholder. We have this problem, whether it's a tenure and promotion committee or academia or whatever.

To me I think an alternative metric for that is how is something being prioritized. If it's needed by another component of the agency and you've been asked to do it, that's a reason for a prioritization. If it is clearly something that is emerging as a new need, and you all feel

that you need to address it, I think how that is prioritized.

And I think it would be informative not only to see how that prioritization is done, but then an outcome. So we have prioritized this because this is needed. And if it is a new technique or a new piece of knowledge, a new AOP, a readout for another part of the agency, then that's something that can be tracked as an impact metric because you've met that need for prioritization.

I think along with that one of the things where I saw some very exciting things and some things that it would have helped me to have heard why this particular approach was chosen is to think about when is a gold standard technique or approach the best one to use, it's the most cost effective, it has a great database you can relate it to, it's going to get you where you need to go faster than anything else. But where are there new techniques that are out there that probably could be incorporated and get you where you need to go, possibly in an even more informative way?

And so I think the choice of some of the assays seem to me, I would have liked to have heard well why are you doing it this way, as opposed to where there are new techniques that are out there, new approaches in the community, why were those considered, were they considered,

and why were they not chosen as the way to go forward. So that type of information would have been very helpful for me.

DR. GANEY: Thank you.

DR. SAUER: Let me emphasize what has already been said. This was a great session. This was probably one of the best sessions that I've sat in for the past couple years. Number one, the divisions have definitely embraced our guidance over the past several years around the format of the presentation, and I think that was great.

And when I think about impact, getting the impact, because that is what it is all about, to be truthful, what I liked during the presentations was when the rationale for the project was really around what the product center needed. That was great to see, because then we saw the need, and hopefully we saw then the solution, and that's the impact we need.

And I know for the division leaders it is tough because what does the SAB do? They want everything, right? We want to see innovation, we want to see impact. But I think it is going to be finding the balance that is really the important part behind the science that's being done. But at FDA it has to be about applied projects in my mind. That's what you're trying to do, you're trying to enhance public health. And I saw that across many of the projects.

And so I think that impact piece needs to be worked out and setup some clear metrics around that. But I think you have all the pieces, to be truthful. I think it is really a discussion about how you weight the different subjects that you talk about during your presentations. Thanks a lot, it was a great couple days.

DR. GANEY: Alex is next.

DR. TROPSHA: So, a great thing to be one of the last to speak among a group of highly distinguished and super smart colleagues is that there is almost nothing left to say. So I want to thank everybody who spoke before me for really capturing a lot of my thoughts.

I agree with Ken about the -- I felt the same energy, and interest in sharing. Maybe it is because people are coming out of the last two years of nonexistence, this is an opportunity to sort of be among colleagues. So I think there are a lot of great things to say, and great presentations.

A couple of thoughts that kind of go back to the issue of impact and innovation. Those are hard to define, I completely agree with Tucker. But what is on my mind is data science. This is something that I think is emerging more and more.

And I think there is a great understanding that there is a universal research currency that is called data

which is associated with every research project conducted within the center, and I am wondering if there are ways of capturing the research data aspect of all projects across the center, quantitating, and realizing that research data streams are associated with research projects as a separate entity and separate currency.

And kind of using this in order to measure impact innovation as in protocols discovered within the center versus outside and implemented in some ingenious way within the center. Collaborations across the center and with other centers within FDA. It could be measured in sort of joint research output. And I think articulation of those aspects might help with quantitating impact and innovation.

I mentioned in my comments for instance that the databases can be given in their usage outside of the group could be tracked, I think that there are similar metrics that might be established. So this is sort of my overall observation and recommendation, is to incorporate research data management in a modern way into the fabric of the center's activity. Thank you.

DR. GANEY: Greg?

DR. LANZA: First of all, since I started coming, and Donna knows how long that was, but the whole big thing has definitely improved in the quality of the science being done. And at that time one of the things Slikker used to

say is that they wanted to show they had peer reviewed publications as a marker of that quality. And I think that they have exceeded that expectation.

And I do agree that they need to also put some weight on the impact to the different centers they're supporting, and I'll come back to that in a moment. But I think that's actually the biggest job, is the regulatory science to help those guys do their job.

The second thing is that I've been watching the AI develop. Of course, I do a lot of this with imaging, in collaboration with big companies, particularly United Imaging. And I think that it has improved a lot. But I wanted to make out the point that when you look at the distribution of AI within the FDA, its applications, I saw a recent article about 150 different ones, I would say 80 percent is in radiology, and then the next level would be cardiology, and then everything else was a blip on the bar.

So I do think that radiology was doing quite a bit, but maybe NCTR maybe needs to do more on the interaction with the radiology center. And I noticed they were spending a lot of their time on the devices. So I don't know how to make that bridge, but the impact there is quite good.

And I asked the question earlier about that, because in the work we're doing, the clear vision is not

just to accelerate, I work in MR primarily in ultrasound, but not only to simplify, accelerate the things so that you don't have techs, but also to do all the postprocessing inline and generate reports.

And the next step from that is longitudinal healthcare management, which is in other words if you use this treatment what would be the expected effect, that's the path. And so I think there is an opportunity for NCTR to get more involved on that.

And the last thing I wanted to point out, being a cardiologist, and having bias, is that a lot of times it is the same old drugs, like the cardiotoxicity of anthracyclines and stuff. I am skating people down here all the time in our scanner, and they are getting checkpoint inhibitors and all kinds of drugs that are causing cardiomyopathies, as well as long COVID and so forth.

And I think it would be useful for them to maybe expand some of the drugs they're looking at from a toxicity standpoint as we go to more personalized medicines, immunotherapies, and stuff like that. And particularly the major organs that are being affected.

And even on the drugs, I am doing research on proteosome inhibitors and things like this, they have plenty of toxicity. It seems to me that because they have these toxicities people aren't, well they're going with

mostly drug therapies, and then all of a sudden they're into CAR-T because they're in multi drug resistant disease. So especially proteasomal resistance will give you an example of multiple myelomas. So I think there is some room for them to expand beyond the drugs they are studying, and have some projects that are more reflective of what is going on in terms of the use of new drugs. That's all.

DR. GANEY: Thank you very much. Chuck?

DR. KASPAR: Thanks to all the presenters. I found all of the talks informative. And I will try not to be redundant in my comments on what other SAB members have already alluded to or talked about. Particularly when it relates to assessments, this was a new area this year, but I would encourage as Tucker has already mentioned this working group on assessing new ways, assessment, and project management even on how these various projects impact the goals of FDA.

In the past we have talked about and encouraged collaborations, both within NCTR as well as outside of NCTR and other centers and divisions within the FDA. Certainly that is happening, and that was very evident in the talks.

Another thing that has been mentioned in the past is reducing redundancies within the presentations. I really noticed that this year. There seemed to be very good

coordination, from the center talks down to the NCTR talks. SO I wanted to mention that.

One comment that has come up in the past, and then I'll bring it up again, is that there is a lot of work to do. There is a lot of work to do, a lot of areas with a lot of limited funds and personnel. And one of the center directors made this in one of his presentations, do you want to accomplish a little on a lot of things, or a lot on a few things.

And I would encourage everyone to re-look at your priorities within the various groups. I guess I'm of the opinion to focusing on a few most important priorities and accomplishing a lot or gaining a lot of ground on our understanding of that particular area. So that's one thing I would point out.

Another area we have talked about over the years, and it continues, it was mentioned in almost nearly every talk, is filling slots, personnel, and in the past we've talked about recruitment. And from what I heard it seems like you're moving on recruitment, I would continue to expand on that. But maybe also look at retention. I know there were some retirements mentioned.

And I'm sure you already do this, but when someone does leave, are you collecting that information. Why did they leave, was it strictly salary. I would guess

it is not always salary. That is certainly a primary driver. But you might be able to get some other information on what is leading to people leaving.

And one topic that got brought up, I think on Dr. Foley's talk, is perhaps center driven projects versus PI driven projects, and is there a frustration there with the scientists in not being able to pursue some of their ideas or some of their interests.

I don't know the answer, maybe you have some of this information already. But I would encourage the center and groups as a whole to perhaps look at some of that information collected on people departing, and focus both on retention and recruitment. And I'm sure you're doing both, but I just thought I would point that out or mention it.

DR. GANEY: Thank you. I don't have a lot to add to what everyone has already said. I agree with what everyone has said. This has really been a really good meeting. And I too for some reason, it's funny that you would say this, I couldn't put my finger on why I thought it was an improvement over previous years, I want to just touch on a few things. Actually a couple that Chuck just mentioned.

One is the perennial problem of filling positions and keeping people working in Little Rock. What I might

just add to what he said is when someone does accept a position it may be good to query what were the selling points, why did they make that decision. If they had other offers, what was it that caused them to choose to come to NCTR. And then that gives you something at least maybe that you could build on for trying to attract people in the future.

The other thing is that I agree that the metrics for success, there needs to be some attention paid to that. And when I look at the center as a whole, I wonder whether it can be the same within each division, or whether there might have to be some tailoring to each of the divisions for what those metrics for success would be. I'm not the one to answer that question. Tucker, you're in a better position to take a look at that. But I would think that that would be something worth considering.

And then finally the issue of prioritization came up within several of the division directors' talks. This also seems to be a perennial discussion that we have. And I will tell you that I remain confused about this, about how projects are prioritized, especially when I think about the objective or the mission or the goals of the center in general, one of them, one of the primary ones is to provide support for the other centers.

Then if there is a project that is initiated within the center, say for example one that the Division of Biochemical Toxicology decides is going to be a really cool project, does that automatically get put on a back burner because it hasn't been initiated by one of the other centers that you're supposed to be supporting. And so that I think there is maybe it's just my confusion, or maybe there's an innate angst there, or at least some obstacles there for setting up ways to prioritize projects.

And it becomes an even more important question in the face of you mentioned that your funding has been reduced, and some of the division directors mentioned that their funding has been reduced, and if you have fewer people to do projects that is going to limit which ones you can do, how many you can do.

So I think that these are all things that require, incidence, though you can look at the prioritizations separately from the metrics. I think you have to think of it as a big bucket, and you have to include hiring and retaining people as well. It's all part of one bigger issue. And it looks like Ken has raised his hand, so I'll stop talking and leg Ken talk, and then Greg looks like he has something that he wants to add.

DR. RAMOS: A quick comment Patti, just to echo what you just stated. One of the notes that I took is that

at some point I think we need to discuss how priorities are set at the NCTR, because a lot of the alignment with impact, and a lot of the alignment with even the way in which divisions are reviewed ought to be framed around how these priorities are established. So I echo exactly what you just said.

DR. LANZA: The one thing that I heard over and over, of course we experienced as well, is the issue of service agreement, especially for mass specs and so forth. And I don't understand why the government, the NCTR as part of the government cannot have preferred vendors to negotiate much better contracts for supporting it government-wide, rather than individual buyers within NCTR having to pay.

I know what they're talking about, it's a $25,000-$30,000 annual service agreement plus other costs. And if you have a lot of them that's a lot of money. So it seems to me that that would actually put more money into the research they're trying to do and less money into the just maintenance cost of keeping the equipment going. And also probably when the equipment goes down it goes down for a while.

So I don't know how that has to be negotiated, or why it hasn't been, but it seems to me that it is time for a more collective bargaining agreement with regards to

these large pieces of equipment that have high annual maintenance costs.

DR. GANEY: I'm actually glad you brought that up because that was one thing that I picked up on as well. I don't know the breadth of the equipment about which you're talking, but I wondered if for some of your equipment, maybe not the really high end technical ones, but some of the equipment you considered hiring someone that you send to courses to learn how to repair and maintain some of your other equipment, and then you would just have one salary to pay, other than 15, 20, 30 maintenance contracts.

DR. LANZA: So how I do it, in our lab, and many of our colleagues, we do that. We are willing to pay $5000 for a service agreement, someone to come and do and fix something. But on some of this equipment just having a maintenance agreement, on a lot of it, I would say equipment that starts running like HPLCs and above, or any of these DLS, $60,000-$80,000, and mass spec and like that you're talking $150,000-$200,000, they need to negotiate. They have a lot of that.

By the way, a lot of this stuff is proprietary. So I had a particular example recently, $160,000 IBIS optical imaging system, and a power supply broke, and we wanted them to come and fix it. They wanted $120,000 to

come do a servicing agreement on a machine that's not worth that much anymore.

And then we worked through it and found that the power supply could be bought from the guy that sells it to them with the cable for under $6000. So we bought it and did it. So there is merit in what you say. But a lot of times you don't know how to fix it because they don't tell you.

DR. GANEY: Tucker, do you want to respond? Or any of the division directors, do you want to chime in here? Just raise your hand, and I will tell you when you can unmute yourselves. Tucker, you are on.

DR. PATTERSON: I will mention that the service contract issue, a lot of that, we do have on-site staff here that do repair our equipment. I mean, we have an on-site contract staff that has a lot of technical expertise. The problem is with the most recent purchases have been extremely high-dollar equipment, and as you mention, Greg, some of these things are proprietary, so the company has to come in and update your software, you can't do it yourself.

Also, it is kind of a two-edged sword with these overarching service agreements. I got into that when I was in the RCRM staff. Headquarters said oh we have this service agreement now, you can have all this safety stuff done, just write a task order, it's going to save you

money. And I started looking at it, and of course those were DC prices, not Arkansas prices. When it's coming out of your budget and it cost you $300 to get a biological safety cabinet recertified, and I was getting it for $125 here in Arkansas with my own contract. There is good and bad about these overarching service agreements across an agency.

DR. GANEY: Any other comments or responses from anyone within the center?

DR. GAMBOA: So expanding on what Tucker just indicated, this is a really complex affair. It is very true that this is an increasing burden and challenge at NCTR to upkeep these instruments. Typically, when you buy an instrument like a mass spec or an imaging instrument, you're looking at somewhere around 10 percent of the initial purchase cost in maintenance contracts per year.

The challenge again as Tucker touched upon is that the number of these things, yes we can fix on site because we have really superb technical support staff through contract already on site. As an example, one of my mass specs, the vacuum pump failed recently. If we were to purchase a new one from the vendor it would be around $9000, it cost us $200 in a new controller for the board, so we can do that.

But we quickly get into more complicated affairs, which has affected in the past when you used to buy these instrumentation, you had access to blueprints and you had access to what was inside. Now you buy black boxes. Most matters are proprietary, they don't share it with us. And at the end of the day sometimes the parts don't really exist in the open market. And if you have to buy from them they may or may not even sell it.

So a really complicated affair, but certainly one that we don't benefit from the flexibilities that academia can enjoy in some of these regards, where some laboratories are like showcases for the vendor, we cannot afford to do that. Actually, I think that you would be surprised at the complexities that we even have to go through to simply establish a maintenance contract. So very complex but very good points that you raised.

DR. GANEY: John?

DR. TALPOS: I just want to make a quick comment about project prioritization, at least within my own division. For me a lot of it comes down to what is the data gap, is there a data gap that we can form an experiment around and actually answer that question. So what I've been trying to get the division to shift from over the last couple of years is doing science in an area that is of

importance to the agency, to doing science that is going to actually answer a question from the regulators.

So for a long time we did a lot of work around perinatal anesthesia. It is still an area where we do some work. But the questions that need to be answered are getting increasingly complicated to answer the still existing data gaps. So because of that, that is an area that we have moved away from a little bit, just because those studies are now becoming higher, and there's an area where we can potentially make a bigger impact.

Now, in some instances these data gaps are coming at us kind of high up at the institutional level. You can think about the work with heavy metals for instance. That was a real big priority for CFSAN. In other instances, they come from communications with the individual reviewers. That's where the ketamine project came from because they were having specific issues around this one problem.

When it is these reviewer driven data gaps, this is an area where we could really value having greater communication with the boots on the ground if you will at the agencies, because they're the ones that know their individual problems, they may not realize that the NCTR can offer experimental solutions. So that is an area where we do need to work to improve communication to help identify those data gaps and actually generate the data that's going

to help them relieve concerns, even if it might not result in a change in a guidance.

DR. EPPIHIMER: So John, in CDRH I want to caution about going to the reviewers themselves. Because when I came here two years ago, there were definitely collaborations on things, even critical paths that were being submitted by my team that said oh, this reviewer thinks it's important, it's a gap. When I went in and actually reached out to the OHT management team, they said this isn't a priority at all for us. This is an interest of that one individual reviewer who, to be honest, wants a connection to research in most cases.

So what we found was, and I could tell you that with even, I'll be blunt, the things that NCTR has done with us in the past are not priorities of the center, they've reached out to a collaborator whether in my group or that, but my staff wanted to collaborate with somebody, but so they just wanted a research project so that they could have a publication.

That's what the impetus was. They were not working on center priorities, or giving the accurate information to NCTR, because they don't know necessarily what the priorities are at the center level. And because they look at it through blinders of their own individual interest in many cases.

So I want to caution at least in CDRH, from my experience, that I've been here for two years, and having to kill so many projects that were not center focused but were more one-off reviewer type things. Because all of our internal proposals, you need to have somebody from another center kind of be on it, like for OCS and stuff like that. My people are more than happy to say they will serve as a collaborator, even though it's not a center priority, just because it's an OCS requirement to have multiple centers on it. So I want to be careful about going to reviewers.

DR. TALPOS: Another point that we struggled with a little bit is of course it is the squeakiest wheel that gets the grease. We do sometimes have that exact same issue, but it does seem that there are some projects that are important, but we have folks that are a little bit lower that are the ones that are ultimately advocating for them.

DR. EPPIHIMER: There are stakeholders that you should listen to. But it comes back to stakeholder input. No project should be started based on input, based on one input wanting it. It should be a collective assessment of multiple stakeholders, to be honest, and management is responsible for center level priorities, to be honest. Successfully ensuring that resources are there to make them successful.

If we don't, again, speaking, what we've learned is speaking to a scientist or just one reviewer, we have found out that 90 percent of the things that get recommended to us from that are not center level priorities.

When it comes to where resources are, we are working with very limited resources, so it is extremely important that to get the full support that we've realized, because again what I keep going back to is the methods, the things that we do, one single reviewer, or a single person at a lower level is not going to make it get adopted by a division, and say on the review side in CDRH. It is going to need that one of the OHT managements to kind of foresee that.

And so I think speaking to the right stakeholders for NCTR would be very imperative for making sure that the work has greater impact and alignment with the center. That is just, again, coming from this and having to do a triage in every program, in every project in my division. And what we've learned in OSEL(?) is that the stakeholder analysis and feedback has to be more than one single person. That may be the impetus for an idea, but it needs to be substantiated with deeper stakeholder feedback.

DR. GANEY: I see a lot of hands raised, so I am going to move on right now. Laura, you have had yours up the longest, so you are on.

DR. SCHNACKENBERG: I just want to respond to Michael. I completely agree. Certainly in the past we have had the case where we've had projects that were not necessarily the pet project, but an interest to a particular reviewer, leadership didn't necessarily know what was going on.

So I just want to say that from a division level we are trying better to just reach out to the leadership at the various product centers. And then from there, if there is a gap or something that we can help with, going to them first and then saying who should we collaborate with on that.

So we are trying to make that effort that you're talking about, where we're starting with leadership and moving down rather than just trying to find an idea that fits within one particular reviewer that may not be the center priority. I will let, since there is a number of hands --

DR. EPPIHIMER: I will agree with that, Laura. That is why we have made all of our gaps public for everybody, so that they are aware of what are management's priorities at the center level.

DR. FITZPATRICK: We also had this problem a few years ago, and it really wasn't fair to NCTR researchers that started on a project initiated by one person and coming back to the center, they're going what is this. So we do have a really good system with NCTR now where they come through our office and the Office of the Center Director. They send protocols to us, we approve them and get people involved in it. I think it has been a really great partnership.

And if we want something done we usually go to Tucker or Gonzalo to talk about it before it goes down to one of the centers. We can't catch everybody, but we are trying to catch any kind of rogue investigators that go on their own. You know how that is. But I think NCTR has been very cooperative. Of course they do great research for us.

DR. GANEY: Alex, do you want to add something?

DR. TROPSHA: I really think the discussion that is going on right now is very important. I don't remember us having a similar level of discussion last year. I want specifically to defend Mike for being really candid about the issue. But the issue, and I hope that the discussion was provoked by the commenters by SAB members, and that is the theme I think that we have reflected upon, which is how projects are selected and prioritized.

And I think this is just sort of the intensity of the discussion right now, and the agreement between FDA people that that's an issue, I think that that indicates that there is something to be done at all levels. And what goes through my mind is, and that is back to Mike's comment that people initiate collaborations but that are not necessarily in the interest of the entire division, and projects that we have to kill.

I think that is an important issue to address as to how the project is selected, what are the criteria for the decision to go on or not to go on. I think it should be a place for investigator-initiated projects because that's what people are excited about, and I think that those should be looked at carefully before deciding to allow or disallow.

But I think there is an issue of discussing project priorities at the level of center directors, if each center has their own priorities, if there is a collaboration then it should be aligned.

And then again back to the research data, research data have to be generated of value to both centers, but within the center the question remains how individual projects at the level of divisions are selected, prioritized, supported. And also I just keep thinking, I

don't think we discussed this a lot, how the projects are terminated.

I was intrigued by what Mike said about terminating a bunch of projects in the last two years. I don't think we talk about criteria for project termination. And so I think these are the really serious and important questions that should be addressed, or probably we should be making those comments in our review as SAB.

DR. GANEY: Actually this is an important thing that we didn't get to discuss. Steve?

DR. FOLEY: On the comment about prioritization, one of the things that is needed in the reviews and approval processes is supervisors from collaborators to sign off on those, and so there are some checks and balances already in there to make sure that the one off that the investigator and other center may have has the manager support. And so I think that is important too.

And at that concept phase a lot of times they can be stopped, and so not a lot of extra time is wasted in developing the full protocol doing that if there's not the support from the other centers, and so I think that has been beneficial in this role as the Director, to be able to understand what you hear as a priority from an individual in the center may not be the center priority, and then that

can help make some of those decisions on what to go forward with and what not to.

DR. GANEY: Does anyone want to comment on the process by which projects are terminated or sunset or whatever you want to call that, or just abandoned? I know John mentioned the possibility of having to do that on some project, but does anyone else care to speak about whether there's a process for that, or it's just one day you wake up and ugh the data are crap, or we're not doing it anymore. I've been there.

DR. FOLEY: A lot of times the projects run their course, because when they're approved they have a timeline, and that sort of stuff. And so they will run their course to hit the metrics, and then when the metrics are hit then they will sunset out. Then there are other times, there are things where we have had to put things on hold.

And I know with COVID for example we had some projects that were being worked out that we had to say all right, we were going to put those on a pause and either not go forward with those at this point, or they were approved concept and not move forward with the full protocol because we've got these other priorities. And sometimes you get down the path, and there is a termination process that we go through in our protocol tracking system if they need to be ended early for a variety of reasons.

DR. EPPIHIMER: Patti and Steve, we are more than happy to share, we have developed, we have defined work instructions because it is part of our quality management system to be compliant with ISO 9001, you need to gather stakeholder input. Your customer feedback is critical as part of that quality management system requirement that we're instituting. We have developed work instructions as part of our quality management system that governs how to analyze stakeholder input and how to prioritize it.

And so essentially it gets designed a score that management looks at the data and says okay, does it meet something that we should invest, and it comes down to you may have three important projects, but you only have resources to do two of them in a reasonable amount of time. You could do all three, but it would take five years to do it, which means that we're not getting impact and value back to the customer soon enough.

So in the end tough decisions have to be made, and okay, maybe we're only going to do one project, or two. But that is kind of discussed and all well-defined within criteria of selection. We have it for sunsetting projects, we have it for things that are interesting, but maybe you need a little more data to say it's worthwhile doing it. So we are more than happy to share all of our work instructions, SOPs around that.

DR. GANEY: Thank you. Greg, you had your hand raised. And then after you speak, Gonzalo you can say your piece.

DR. LANZA: The only thing, I'm listening to this, and I spent a lot of years in industry. We used priority research budgeting every year. And so we built our towers with the priorities that the fundamental ones would be at the bottom, and then you get this more differential choicing thing, and it was very formal. I think that, I am not clear, is that going on in each of the groups here, or is that something that each person takes their own thing? I was at Monsanto at the time, when there was a Monsanto, but essentially every place had priority research budgeting for every budget. Does that go on?

DR. GANEY: Does someone want to answer that question, or should we allow Gonzalo to take that one, since he had his hand up anyway?

DR. PATTERSON: I can speak to the prioritization. Of course we have prioritization projects. We can get in a little more detail on that in the closed session, I can tell you what drives a lot of that. But as I mentioned before many times, we have money that comes in on our VA that is earmarked for a particular type of work. That is congressionally mandated, we have to do that type of work with the money that we receive.

Of course those are going to be going to the top of the list, because then we have to turn around when they have a data call and say okay, how did you spend this money, we have to show them the projects that we spent the funds on. So yes, we definitely prioritize that.

This past year of course the Predictive Tox Roadmap Funds, that was literally the majority of the VA discretionary funds that we received internally to distribute for research. So everything has to focus in that particular area with using those funds.

DR. LANZA: That is exactly what I mean by priority. That is at the bottom of the tower, the foundation of the tower, and then you move up things that are older and less start to fall off automatically in the process.

DR. GANEY: Thank you. Gonzalo?

DR. GAMBOA: I just want to bring a little bit of nuance to the discussion for everything. Certainly the NCTR exists in a context, it is a regulatory context, and our main charge is to assist the product centers into discharging their regulatory duties. By and large the vast majority of the work, the protocols that we have have an identifiable regulatory endpoint, or informational value.

Having said that, we also need to realize that we have the staff, we have a body of scientists that need to

keep their mind open to what's going on out there, to new technologies, to new approaches.

Sometimes we need to allow them to pursue certain research ventures that do not necessarily have a discrete regulatory deliverable, but that at the end of the day, allowing them to do this to explore any platform for example in the absence of specific regulatory context can actually enable them to attend to future needs better.

One of the points that is perhaps not entirely clear to all of us here is that very often NCTR needs to attend to emergencies from the product centers. So there is research that we can see into your eyes and we know that it is going to pop up.

But very often, 7:00 PM and I get a phone call say from Suzie, and Suzie says okay, we have a problem, let's discuss this. And we do our best to tend to the needs of the product centers. But in order for us to do that we need to have a little bit of capacity to be ready and to understand new tools. So what I wanted to bring here is that you should not really expect NCTR to always be able to justify an immediate regulatory outcome for all possible research products that we have.

DR. GANEY: Are there any other comments? This has been a fairly robust discussion for Zoom. If there are no other comments, perhaps we will end the public portion of

the session. I will thank everybody who participated, all of the presenters, all of the SAB board members and everyone else, and our IT folks, thank you. And we will end this.

And then SAB members and Tucker and Donna will join on a private call, you should have gotten an email from Donna about the new login. We will do that. So it's 12:15 now, let's just end this one and start anew. So I'll see the rest of the SAB members and Tucker and Donna in a few minutes.

(Whereupon the meeting was adjourned at 12:15 p.m.)