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FOOD AND DRUG ADMINISTRATION

CENTER FOR BIOLOGICS EVALUATION AND RESEARCH

AND

THE WILLIAMSBURG BIOPROCESSING FOUNDATION

ADENOVIRAL VECTOR SAFETY

PUBLIC MEETING AND WORKSHOP

Thursday, February 1, 2001

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P R O C E E D I N G S

MR. CARSON: Well, good morning. My name is Keith Carson and I am the chairman of the Williamsburg Bioprocessing Foundation. We have co-sponsored this meeting with CBER, as part of FDA. I will be moderating the morning session, so I will be the one keeping track of the time and starting and stopping speakers, and the question periods. The format for the meeting--there are two sessions. This morning session, which will go until noon, is intended to be a public meeting, again, open to the public, basically, to advise the public of ideas, plans, suggestions for developing a voluntary standard for adenoviral vectors and to allow public comment and questions.

This afternoon's meeting is starting at 1:00 and, as you will see, we put a map and we did not give you directions, but we gave you the location and a map--it will be in Building 29B, on the other side of the world, the other side of the campus. That meeting is primarily intended for the adenoviral standards working group which has been established, even though, certainly--seating will be limited for that meeting. The space is limited, but

certainly those who want to participate to the extent of observing and commenting are welcome to do so.

Each talk this morning will last approximately 20 minutes and will be followed by a 10 minute period in which anyone can ask questions. Anyone who does ask a question please stand up, speak clearly. If you cannot be heard, we will ask you to use the floor microphone that is provided. We will ask you to identify yourself before you ask the question. The morning meeting is being transcribed. The transcription of this will be made available within ten working days through the Freedom of Information Act, and facilities available through the documents management branch here, and also it will be posted on both the WBF and the CBER websites.

Copies of all the talks are in your hand-out, along with bios and abstracts for each speaker. Also, I wanted to note that there is a reprint or a copy of an article from "Molecular Therapy" behind the presentations that was copied with the permission of the American Society for Gene Therapy.

I would like to bring up our first speaker, Steven Bauer, who is a senior investigator in the Division of Cellular and Gene Therapy in CBER.

FDA RECOMMENDATIONS FOR ADENOVIRAL VECTOR CHARACTERIZATION

DR. BAUER: Thank you very much. Is this microphone working? Thanks. It is a privilege to be here today, and I want to first start off by expressing my thanks to the Williamsburg Bioprocessing Foundation, Keith and his staff, for their participation, co-sponsoring this meeting, allowing us to get together to talk about this initiative to develop an adenovirus standard. We are very happy that this initiative is taking place and also want to acknowledge the role of the people in the gene therapy community who are gathering here today and have gathered back in October to help make this become a possibility.

You will hear a lot more later today from our next two speakers, Stephanie Simek and Beth Hutchins, about the history of this initiative, how it came about and what the details of it are. I thought in my talk this morning I would talk a little bit about how we decide to characterize different products that come in, how the IND regulatory

process works, and how this initiative to have a reference standard that everybody can use will fit into what we hope are improvements in our ability to quality control vector products. That is my title, "Where We Are and Where We're Going."

As I said, I am going to talk about how we review the products, the process and the goals of that review process, what the current criteria for adenovirus vectors are, how these come about, and talk about where the standard will help improve the process. So to give broad brush strokes about how this works, starting from very early interactions with the agency, pre-IND meetings, where we give advice about what kind of characterization, safety testing, clinical trial design should be done through the different phases of the IND, down to product licensure. Characterization of products is a very important component of ensuring the safety and the efficacy of new products, and we think that having a reference standard will improve our ability to assess products at each of the stages.

So, how do we actually look at a product when it comes in the door? We have somebody interested in trying a new gene transfer approach, and we will restrict our

comments mostly to adenovirus. We have people in house who look at the product, the manufacturing and the quality control and we look at both the final product and the process of getting there, and that is an important concept in biologics oversight. The process is important because you're dealing with biological systems which are inherently complex.

We also have people who look at preclinical data, and I did not finish this bullet point, but safety before exposure of human beings to a product, and also look at the rationale and potential efficacy of preclinical studies as a way to support the idea of justifying trying this new product in people.

We also have people look at the clinical trial design, it's safety parameters, how it will be monitored, and an important process, informed consent. I think having a standard will impact all of these areas through better characterization of products that will allow us to better assess the preclinical data and help to determine or judge the safety before hand.

So, how is vector quality assessed? The goal is, first and foremost, safety, but purity and potency are also

important and I will discuss how having a good reference standard will improve all of these parameters. As I said, we look at the process and the product. Really, for production of a vector that is broken into, sort of, two components, a cell substrate, meaning the cells that are used to propagate a vector, and then the virus bank of the vector that is actually going to be used.

In order to qualify and test the cell substrate, we have a system whereby you create a master cell bank that you can go back to and reliably start from the same starting point for many years of product manufacturing. From that, you make what's called a working cell bank. I should stress at this point that these are general approaches. Everything, really, that we review is done on a case-by-case consideration, so, for instance, some people do not actually make working cell banks for adenovirus production, but I will be talking about both of these at any rate. All of the things I'm going to be talking about are not universally true for every single product. So the cell substrate characterization is important and viral bank--this is the actual vector that is going to be used. There is characterization of that. Also manufacturing

ingredients and the very final product--these are all important to look at.

So, this is the kind of scope of what our review is. The way we evolve our considerations of what is important is primarily in-house, through looking at preclinical data, clinical trial data and product review, but it is an interactive process and it is guided by what is required by law, the CFR requirements. We also have other feedback mechanisms whereby we solicit advice or hear advice from a variety of stakeholders in the process.

Reviewer experience is a very important aspect of how we generate the recommendations for what kind of testing is important. That comes through both research projects that people have in-house--in CBER we have a research review model from their scientific experience, and also just from their accumulated wisdom after reviewing many files. We also take particularly thorny issues to FDA advisory committees, and if these are either controversial or hard issues to deal with, we take them to panels of outside experts and they help us to decide what kind of safety testing, what kind of clinical trial design parameters to use.

Just recently, we had an FDA advisory committee that talked about, at least in one part, structural characterization of gene therapy vectors, and I will discuss a little bit more how that--their recommendations about how we characterize vectors. Also, the gene therapy community, from individual sponsors to organizations like ASGT--we have discussions and fora and teaching sessions with them. All these feed back into how we do review of clinical trials.

Also, we have a very important feature, the recombinant DNA Advisory Committee under the Office of Biotechnological Activities. We have gene therapy safety symposia and public review of different protocols. All of that feeds into how we set and change our standards for how gene therapy products are reviewed. Over the past years, in many of these fora we have seen a growing desire to have some way to have comparability between measurements of certain aspects of adenovirus gene therapy products, and this call for a reference standard has been part of this feedback we have received over the years. We're very happy to see this process is going forward.

I explained the parties that are involved and how we evolve criteria for analyzing gene therapy products. Initially, when we first see a new vector class, and actually throughout our experience, everything really starts as a case-by-case analysis of what the vector is, how it is manufactured, what it is going to be used for, who are the patients that will be exposed. Through this kind of process we also develop general guidelines of what kinds of things we need to test, and this happens through meetings of product class reviewers.

We get together within CBER and talk about all the different INDs we're looking at, the RAC, and I already talked about these parameters. But, also another thing is the March 6th gene therapy letter that went out to sponsors of gene therapy trials. That has been a very useful collection of data for us to look at. Well, how are our recommendations for testing being met? What kind of problems are people out there experiencing? Do there need to be changes in some of the criteria or not? These have been very useful tools in looking at how we test products.

Now, I am just going to go over the kinds of things in the rationale for different testings, and this is

not meant to be a real in-depth review of all this, but just to give a general understanding of how we look at these products and where a standard might fit in.

This is the cell bank. The cell bank that you use to propagate the vector is a very important aspect of looking at safety. The things that we look at are very basic for safety, such as sterility--you don't not want any bacteria or fungus growing in the cells, and mycoplasma. An area that receives a lot of focus is adventitious virus. There are both in vitro and in vivo assays for that.

In vitro means taking that virus preparation and looking in tissue culture for viruses that you do not expect to be there. Things like this have grown through experience over the years, where actually it is surprising that sometimes viruses that you don't expect to be in a master cell bank pop up. We feel these are very important characterizations that need to be done, master cell banks.

In vivo viruses are looked at by inoculating animals with different preparations from the cell bank and looking for the presence of unexpected viruses. If certain ancillary products are used, such as fetal bovine serum or porcine trypsin, it becomes necessary to look at viruses

that can come along in those preparations. Looking for the presence of specific human viruses is important, depending on the cell that is used to propagate the virus. Depending on how master cell banks are stored and what history they have had, it might even be important to look at the presence of replication competent adenovirus. This is a place where, and I'll discuss this in more detail, having a reference standard that you can look for the accuracy of your infectious titer assays might be useful.

Thank you. How much time do I have? I stepped on the controls. Okay.

If you expand a working cell bank, sterility, mycoplasma and then some basic characterization of the cells--if you use a working cell bank, these things, morphology, isoenzyme characterization, these help us determine that the cells look like they should and that they are of the origin of the species. It is surprising how often you will find, for instance, that cells from different species are stored and used in facilities where gene therapy vectors are made. So, it is important to say, "Yes, we have not gotten our cells mixed up." And an adventitious virus is to be looked at in vitro assays.

A master virus bank is characterized by some kind of identity. This is just to assure that what you have actually ended up with in your production of a virus bank from which you will start subsequent manufacturing, really is what you intended it to be starting with. Today, we have looked at the sequence of the active inserts, the transgene and flanking regions, and then looked at the rest of it by restriction mapping. There was some discussion of this at the advisory committee meeting of having vectors up to 40 KV completely sequenced, so that is something we're looking at in-house and might change in the near future. The activity is important to show that the transgene you have is actually expressed as protein, and then has some kind activity associated with it.

Finally, the titer is a very important aspect, and this is a place where having a reference standard will be useful. The titer needs to be looked at in terms of both its ability to infect cells and the particle count. Having a reference standard will allow us to say that Manufacturer A, B, and C have used an assay that is tied to a reference standard, so we can have more confidence that there is comparability between preparations of vector. I

will get more into the importance of particle count and the ratio between infectious titer and particle count in a little while.

A master virus bank also needs to be tested for sterility, mycoplasma, adventitious virus, and particularly important here is this replication competent adenovirus. In a stochastic process there seems to be a capability of having recombination between elements, genetic elements, within the cells that are used to propagate adenoviruses, and with some frequency they develop replication competent adenovirus. We struggle constantly with how much of a safety issue that is, so we feel it is very important to be able to quantify that accurately. So, that is a place where having, again, a reference standard with an acknowledged titer will be useful.

And then finally, the final product, again, in terms of safety, sterility, mycoplasma, endotoxin, need to be minimized. The product needs to be sterile. General safety is sometimes asked for, and I think that depends, again, on what the product is being used for. It is not a universal requirement for adenovirus vectors, but this is something we're considering in-house, as well. And

adventitious virus--the in vitro virus assays are expected to be done in final product, adeno-associate virus and then replication competent adenovirus.

Currently, our recommendation is that there be less than one replication competent per 10-to-the-ninth--or infected particles. I misprinted that. That is important--platforming units. This, I think, is an important area where the reference standard will be useful in being, again, able to compare between different manufacturing schemes, different lots of adenovirus within one clinical trial, and allow us to more accurately and precisely monitor the amount of replication competent adenovirus that might be produced during the manufacturing of an adenovirus stock.

Also, and you will hear more about that later, the identity, the activity of the transgene, just as I talked about the potency, is important--I should point out that the potency is different from activity. This is something, for instance, if you have a transgene that should help in tumor killing and you show that it's expressed potency would be going a step beyond in showing reduction in tumor burden, for instance.

Titer is, again, a place where the standard would be very useful, both in terms of having more precise and accurate particle counts and being able to measure infectivity, and then to be able to determine the ratio of these. Currently, our recommendation are that there--this ratio should be less than 100. The concern here is that you don't want to have a mixture of proteins that are potentially mediated inflammation, or other toxic adverse events with very little chance of actually having an infectious particle that will really give you the effect that you are looking for. So, this is an extremely important place to be able to measure accurately and precisely both the particle count and the infectious particle.

Finally, purity is important in terms of, do you have DNA left over from the cell substrate or protein. What I have done is just give you broad brush strokes and rationale for how we get to the recommendations we have for characterizing these products and how we think an adenovirus reference standard that everybody can use will help improve the precision of titers, both in terms of particle, infectious and RC assays. We're convinced this

will lead to an improvement in safety and also our ability to assess the efficacy of these products, to be able to more closely control critical doses, so we can tie safety and dose response to the titer of these products. It will lead to increase comparability between clinical trials, and also it should benefit our ability to look at preclinical studies that have used well-characterized, well titered adenovirus.

So with that I think I will stop. Thank you for your attention.

MR. CARSON: Now, we will take approximately up to 10 minutes for questions.

FLOOR QUESTION: I was wondering whether CBER was going to issue guidelines on safety testing and general testing of adenovirus vectors.

DR. BAUER: We're working now towards updating the gene therapy guidance, and we're working towards having, not only for adenovirus, but all gene therapy products, kind of an update on what kind of characterization we would like to see. I do not really have a good timeline for that, but we're working on that in-house right now.

MR. CARSON: I'm sorry. Since we're doing a transcript I understand that to get it on tape we have to use the floor microphone. Please, if you don't mind, if everyone would use the floor microphone.

FLOOR QUESTION: Can you clarify the general safety? I think you said that this is not a universal requirement. Is this only for gene therapy products or with any product made with gene therapy vector?

DR. BAUER: Yes, with adenovirus we have not been universally requiring that, but I think that is something we're reviewing in-house right now, whether or not we should.

FLOOR QUESTION: Can you clarify whether this is just the gene therapy CBER requirement or it also applies to the vaccine?

DR. BAUER: I do not think it applies to vaccines, but I don't--

DR. SIMEK: It still is required for licensure for gene therapy, we're just not requiring it in Phase I at this point--it does not apply to vaccines, but it safe to say it applies to gene therapy.

DR. BAUER: That is an important clarification. For Phase I, general safety is not required, but by licensure it would be. Thank you. There is a question over here.

FLOOR QUESTION: I had a question on the slide that listed master cell bank safety and you referred to RCA. I was trying to understand what you would be looking for in a cell bank with respect to RCA.

DR. BAUER: If you, for instance, have a cell bank that might have been exposed to some vector in the past, and there is some possibility that you might be propagating that virus in the cell bank and not necessarily from your product, that is something we would like to know about. That is more of a case-by-case consideration, but it is something we thought is a possibility to look at.

FLOOR QUESTION: So, you're just looking for a contamination with adenovirus.

DR. BAUER: Right, and it could be another vector and its RCA or adenovirus, wild-type.

FLOOR QUESTION: The detailed procedures in testing the adenovirus, will they be available and will they be standardized? For example, when you say testing

RCA is a different matter and testing contamination by viruses is also a different matter, different labs using different approaches. Will those be all standardized and will there become a printed material available?

DR. BAUER: Right now we don't have standardized methods for many of these assays unless they are, for instance, from the CFR, and that is to allow flexibility in people to meet these recommendations. Over time, it might be that, for instance, the working group decides there is an SOP that would give you the most confidence in a certain preparation, and it's your ability to titer it, but that is not something we are requiring at this point. For most of these recommendations it's, you know, here is the specification we set and the goal, but then there is a lot of flexibility in how you meet that goal.

MR. CARSON: There will also be a panel from 11:30 to 12:00, so you will have a chance to ask each one of the speakers additional questions. I would like to bring up Stephanie Simek, who is now a product reviewer, probably has always been a product reviewer within CBER.

FDA PERSPECTIVE ON THE DEVELOPMENT OF AN ADENOVIRAL STANDARD

DR. SIMEK: Good morning. I would like to thank everyone for attending this public meeting. Thank you for attending this meeting today and giving us the opportunity to discuss with you some of our initiatives in collaboration with industry and academia to develop an adenoviral standard. I would also like to discuss what we believe to be the importance of developing the standard and also hopefully what will be accomplished by having a standard in place and available for use by those doing adenoviral gene transfer studies.

First, I would like to give a brief background or history on the events and efforts that have taken place and ultimately lead to this current initiative, which is the development of an adenoviral standard. In the early 1990s, adenoviral vectors were initially used for the treatment of cystic fibrosis. After the initiation of these clinical trials the cystic fibrosis foundation openly discussed the need for an adenoviral standard. Over the ensuing years there were multiple discussions relating to the development of the standard, but during this time there was no real

consensus reached as to what would serve as an appropriate standard.

In 1999, after the tragic death of Jesse Gelsinger [ph.], NIH,OBA requested that investigators and sponsors submit all relevant preclinical and clinical data regarding adenoviral vector safety, toxicity and efficacy. In addition, NIH,OBA established a working group, the Adenoviral Vector Safety and Toxicity Working Group, whose mission was to conduct and in depth review and evaluation of all the safety and toxicity data that had been submitted. Also at the same time, there was a safety symposium held where investigators met to discuss both preclinical and clinical adenoviral studies.

As will be discussed in more detail in the following presentation by Dr. Beth Hutchins, on October 5, 2000, the Williamsburg Bioprocessing Foundation held a meeting in Washington D.C. to discuss the feasibility of developing an adenoviral standard. Before I discuss some specific issues resulting from the October 5th meeting, I think it is important to mention just a few recommendations that were proposed by the NIH AdSAT working group that directly relates to adenoviral vectors.

The working group recommended that there be development of qualitative and quantitative vector standards, one of which would be a standard for adenovirus. It was agreed that the development of a standard could then be used to determine and compare particle number and infectious titer between different product manufacturers. Also, it was agreed that the development of an adenoviral standard would allow for comparison of toxicities observed in different studies. This would allow for comparison between preclinical studies as well as allow for comparison of toxicities better observed in different clinical trials.

What I would like to do is discuss some of the FDA's perspectives on the importance of developing an adenoviral standard. First, there is the concern over the precision and accuracy of adenoviral titers. Although measurements of virus particle counts are more precise, the current biological assays for infectivity, there are still no consistencies in the values reported because multiple methods are still currently being used to determine virus particle counts.

The most common method currently used is to measure the absorbants at OD260 after lysing the virus

particles and then converting this value to a particle count based on a published extinction coefficient. This method of measurement may not always be consistent because it may actually differ depending on the final formulation of the virus preparation. There is even more concern for measuring infectious units, since it has been reported by numerous investigators that, at best, there is a 30 percent level of imprecision in the biological infectivity assays that are currently used.

As I am sure many of you may be aware, the FDA currently requests that all dosing be based on particle number. Since there's still a considerable amount of inconsistency in how viral particles are measured, it is still quite difficult to compare dosing between different clinical trials. We are also concerned about the sharp threshold of the fact that we observed in the dose toxicity curve. Having this threshold, above which we began to observe increased toxicities, makes it extremely important to have a more accurate means of measuring how much vector load is actually given to subjects.

It is clear that having a more precise and consistent measure of viral particles will ultimately lead

to consistency in clinical dosing. This means that it would provide for better dose control, which means a closer approach to the maximum tolerated dose, smaller dose increments, and also provide a better means for analysis of dose-related adverse event.

The agency also has safety concerns relating to the use of adenoviral vectors in clinical trials. It has always been a concern as to the level of replication competent adenovirus that is present in each dose of vector that's given to subjects and actually how much RCA is really safe. Currently there are also inconsistencies in what analytical methods are used to quantitate RCA, so, therefore, the RCA testing is not consistent among different manufacturers. Until there is more consistent data available concerning the level of RCA actually given to subjects, the agency has taken a more conservative approach and assumes that there is a safety risk. We also have safety concerns regarding the actual amount of vector particle that is given to subjects at higher doses.

Our approach to standard development requires that a standard be well tested and characterized, and serve as a testing reagent to be used as a reference to

standardize both physical and biochemical measurements. In the case of an adenoviral standard, it should serve as a measurement for both virus particles and infectivity. A standard should also lead to development of SOPs that can be used by investigators and manufacturers to validate their own internal testing procedures. The Office of Therapeutics has some previous experience with development of a standard for replication competent retrovirus. This standard was developed as a collaboration with the FDA under the direction of Dr. Carolyn Wilson, in collaboration with ATCC, industry and academia.

Now what I would like to do is discuss some of the FDA's initiatives relating to the development of an adenoviral standard and then I would also like to introduce some new research that will be starting in the Office of Therapeutics, under a new investigator, Dr. Andrew Burns. First, I'm going to describe a collaboration that has been initiated with industry and academia and, specifically, I will discuss the FDA's participation in a working group whose mission will be to oversee the development of a standard. Dr. Beth Hutchins will describe in more detail

the actual role of the working group in the next presentation.

As I mentioned in the beginning of my talk, there was a one day meeting held on October 5th in Washington D.C. This meeting was organized by the Williamsburg Bioprocessing Foundation in conjunction with the FDA, industry, academia and other regulatory agencies. The purpose of the meeting was to discuss the need for an adenoviral standard. The overall consensus of this meeting was to develop a well-characterized standard, and it was also agreed at this meeting that the FDA would take the lead in this initiative using a working group approach. The working group will be responsible for selecting an appropriate group or groups to manufacture, characterize and distribute the standard. Again, Dr. Hutchins will describe in more detail the actual duties of the working group in a few minutes, but I think it is important to state here that it was clear from this meeting that industry, academia, as well as the FDA, are very committed to developing this standard and it was clear from our interaction with individuals present at this meeting that this was truly going to be a collaborative venture.

Before I go on and discuss the FDA's role in the working group I think it is important to mention that the FDA has a history of collaborating with the external scientific community. These collaborations have been done on a more formal basis through cooperative agreements such as memoranda of understanding, co-sponsorships, partnerships and contracts. The FDA has develop the leveraging initiative whereby we can invest our resources in collaboration with others outside of CBER to meet our public health responsibilities. This strategy allows for maximum flexibility and it allows the job to get done faster and with expertise that we may not always have in-house. By pooling our financial and intellectual assets we're able to achieve results greater than any of the participating organizations may be able to achieve alone. For those of you who are interested in obtaining more information on the FDA's leveraging initiative I've included our website at the bottom of the slide.

As an example of leveraging agreements there was a co-sponsorship agreement signed between the Williamsburg Bioprocessing Foundation and the FDA. This agreement made possible this meeting today and also provides for public

discussion and input. Furthermore, a partnership agreement has been drafted between the Williamsburg Bioprocessing Foundation and the FDA. This agreement allows for the collaboration between WBF, FDA, industry and academia and provides for the participation of the FDA in development of voluntary industry standard. As a partner in this agreement, the FDA will help in identifying relevant criteria in the production and distribution of the standard with the goal of improving our ability to evaluate the safety of adenoviral gene transfer studies.

As I just said, it was agreed at the October 5th meeting that the FDA would take the lead in this initiative using a working group approach. Our role in this working group will be to take the responsibility for leading the process to evaluate and select the group or groups that will be responsible for manufacturing, characterizing and distributing the standard. In the partnership agreement I had just described to you, Williamsburg Bioprocessing Foundation will serve as the facilitating entity for the working group and the FDA.

WBF will post RFAs, announcements and meeting minutes on the website. They will serve as a kind of

clearinghouse for information on standard development. WBF will also oversee the performance of each contractor that is involved in the specific steps of this manufacturing standard. So, part of the FDA's role in the working group will be to review proposals for vector production and then make recommendations back to the working group for selection of a group or groups that are going to be responsible for manufacturing this vector.

The FDA will also take the lead on recommending standard qualifications. And you have just heard in the last presentation by Dr. Bauer what some of these qualifications or testing requirements by the agency are. Once testing of the standard has been done, the FDA will collate the data and then present the results of the testing data back to the working group. Overall, our main function will be to provide guidance to the working group and to ensure that the standard meets our qualifications and will be acceptable to the agency for use as a reference standard.

I have included this slide for those of you who may not be familiar with the overall process involved in manufacturing an adenoviral standard. For those of you in

the audience who are, I apologize. This is extremely crude, but I think it is important to describe to those not familiar with this process the steps that are involved in manufacturing a lot of adenovirus, because these are many of the similar steps that will be involved in manufacturing a standard itself.

First you have the production of the master cell bank. As you just heard from Dr. Bauer, the master cell bank needs to be tested for safety and this includes test such as sterility, mycoplasma and freedom from adventitious ages. It also needs to be characterized. In this case, the master cell bank will be of human origin. Routinely, 293 cells have been use to generate the majority of adenoviral vector products, but it was decided by the working group that in this case it does not necessarily have to be 293 cells, but it needs to be another cell line that is routinely used for adenoviral production.

Next is the generation of the virus itself. The consensus of the group was to use a wild-type Ad5 virus, and the reason being that Ad5 is the most commonly used backbone for vectors used in gene therapy clinical trials and also the vector is infectious and would better serve as

a standard for infectivity essays. It also removes the issue of contamination with wild-type, which would be an issue if defective vector were used as a standard. I should mention here that there was some concern for the use of a wild-type virus being introduced into some facilities. Because of this, the working group did decide that a defective vector may also be produced at a future date.

Next in this scheme is the production of the master viral seed stock, or can be referred to as the master viral bank. Again, you have heard a bit about this in the last presentation by Dr. Bauer. The master viral seed stock also needs to be tested for safety and shown to be free of human pathogens. Then the seed stock is used for the actual generation of the production of the vector lot. This lot will also need to be characterized, as an example, for purity and also tested for safety.

Lastly, after the lot has been tested and released, it will need to be vialed, frozen and stored. Then the frozen vials will need to be tested for stability to ensure an expiration date, establish an expiration date.

I would just like to briefly mention that we have a new investigator, Dr. Andrew Burns, who has just joined

the Office of Therapeutics in the Division of Cellular and Gene Therapy. Andrew brings with him expertise in adenoviral research. His research will be focused on human and murine adenoviral interactions with viral receptors. He will also be studying the effect of receptor interactions on viral tropism and pathogenesis.

I would like to end here today with the slide describing what we believe will be accomplished by developing an adenoviral standard. First, it will mean the production of more consistent, safer and higher quality adenoviral vectors. The standard will also allow for comparability between preclinical studies, as well as allow for comparability between clinical protocols. Lastly, having a standard in place will ultimately lead to the development of regulatory policies. Thank you. Questions? Okay.

FLOOR QUESTION: I just wanted to ask about your last slide here. When you talk about allowing comparability between pre-clinical and clinical studies, who is doing the comparison? This would allow for FDA comparison across manufacturers or, I mean, who would this assist, really?

DR. SIMEK: I want to be careful how I answer that, not being a clinician. We do not have, although the clinical reviewers do try very often to compare different studies--I think it will both. It will allow, I think, the different manufacturers and sponsors to be able to compare their studies amongst themselves, but for us it will be very important because at present it is very difficult if you look at two different clinical trials to be able to compare the dosing between them. That is very important when you start thinking about adverse events. So it really will be a tool for us as well, but I think it should be able to benefit everyone.

FLOOR QUESTION: I think the question was asked once, but can you further clarify for us the difference between using adenovirus as a vaccine vehicle verses for gene therapy? What is the different requirement between the two?

DR. SIMEK: I am not in a position to discuss what any of the testing requirements are for the Office of Vaccine. I will state again that everything we tell you today pertains to the Office of Therapeutics and, specifically, for the Division of Cellular and Gene

Therapy. Although we all try to harmonize, there are just differences, so testing may be different and, again, this is done on a case-by-case basis. I cannot and will not tell you the similarities or differences between the two. This is just for gene therapy products.

FLOOR QUESTION: I have a follow-up question. Do you have any plans to integrate the gene therapy group and the vaccine group involved with the review of the adenoviral vector based products, vaccines, in the future?

DR. SIMEK: Dr. Zoon has always like to have, in CBER, to have harmonization as much as possible between different offices, and of course we work very hard to try to do that. Again, just by the nature of the products, that may not always be the case. So, as we do try to work toward that, there is nothing I can say today that definitely--what the initiative is or what the result will be.

FLOOR QUESTION: I forgot to mention--the second question is can you tell us more about this working group, how it was chartered, who is participating in this working group and a little bit more about it?

DR. SIMEK: Dr. Hutchins will describe that in a minute. Also, I believe on the Williamsburg website there is a list of who the participants in the working group are. It originated from the October 5th meeting.

MR. CARSON: We are set up for the break. We did finish a little bit early, but let's go ahead and take the break.

[Recess.]

MR. CARSON: As you will see in the agenda, after Dr. Hutchins' talk, we will set up a panel here in the front. Again, that is for open discussion, questions from the audience, and again, all the speakers will be there for you to ask additional questions.

Following the panel, at or about 12:00, we are going to turn you loose for lunch. I have heard two recommendations. There is a cafeteria at the end of this hallway, the other end, that you might try. Also, I understand the one in Building 10 on the second floor, the one on the bottom floor apparently is closed, but Building 10 on the second floor I hear is a good choice. Other than that you are on your own. There is also a cafeteria in Building 31. That is close actually. It's in the opposite

direction. Then you'll notice, again, on your map that--where Building 29B is, and we will start there exactly at 1:00 PM.

One comment about the working group, again. We had several questions. The adenoviral standards working group is a group of volunteers, people who volunteered to serve on this working group. Anyone can join the group. It is open. It is currently about 32 members. We did not publish it in the hand out, but again it does change on a daily basis. It is on the WBF website. There are several references in your hand out on where and how to get to the website. There is actually a map of the website that you can see once you get on there and you can go and check the membership at any time.

With that, I am going to turn things over to Beth Hutchins, who is Director of Process Science at Canji.

WORKING TOWARD AN ADENOVIRAL VECTOR

TESTING STANDARD

DR. HUTCHINS: Thanks, Keith. Can everybody hear me? I am going to, again, just refer just a little bit to how we got where we are with the working group and then talk about the mission of the group more specifically, in

terms of how it functions, and a little bit of the details of the initiative and where we are in the process.

From the industry part of the community side--really, two years ago a group of us started talking about this at one of the Williamsburg Bioprocessing Foundation's conferences, which is on viral vectors and vaccine viral processing, talking about exchanging our ideas on characterization of gene therapy vectors, and started informally sharing information. That became more of a collaboration among some groups and in 1999 we got more specific about that, and then talked about the idea for a conference about characterization of vectors. Right after that was the safety meeting on the adenoviral vectors in conjunction with the December 1999 RAC. Those ideas sort of came together for developing a working group to develop a standard.

Just so you have a better idea of what really happened at the October 5th meeting, it was organized as a collaborative process with Williamsburg Bioprocessing channeling things and FDA, industry and academia participating. We had more than 115 attendees and we had representation from regulatory agencies, not just from this

country, contract testing labs, academia, big Pharma, and small biotech companies, and standard-setting organizations, specifically, the USP, the NIST and NIBSC. The way we handled this meeting was to get perspectives from the different segments of the gene therapy community, and then to look at how standardization organizations normally go about developing a standard, because originally the thought was maybe we should have one of these groups do it. But the timelines these groups tend to use was a little too long, even though we liked the elements of what they did what they did, so it was determined that the working group would be the best way to accomplish this in a more appropriate timeline.

We have talked about technical data that related to these issues and had quite extensive discussions in small groups, and then came back and that's how we ended up developing consensus during this meeting. Again, we were trying to identify technical practical issues and then come to an agreement on how to proceed, and we did. I just reiterate why, at least in the gene therapy community, we think it is time for a standard.

There are a number of the adenoviral vectors that are in later stage clinical trials now, so I don't know exactly when it will happen, but the possibility is getting closer to having an approved product. The safety profile of a new drug and particularly a new class of drugs is going to require careful scrutiny by the agency, looking at both that a product as an individual entity, but as well as what they know now from the class that that product represents. As Stephanie and Steve pointed out, dose is really the key to understanding what is going on both with safety and efficacy and the fact that we have observed clinically that there is a sharp--there is some threshold upon which safety becomes a real issue. Defining that carefully for the adenoviral vector in general and, specifically, for particular products has to require a much more accurate understanding of dose and what impurities in a dose mean.

Currently, comparability between entities is not possible. That is everything from RCA to the actual particle number. That is because you have the toxicity of the particles themselves, which may in fact be the largest single issue, as well as an ill-defined risk that is

associated with RCA. This lack of data forces the agency from a common sense perspective to be more conservative. Developing a standard to make the information or uniform or at least more interpretive across different studies, be they preclinical or clinical, really benefits the entire community.

I won't really go through this, except to say that particle number, while a variety of methods are used, is fairly precise. Particle/infectious titer ratio, which we all use routinely to monitor consistency of batch preparation and to assist in monitoring stability is an important criteria. That is why in the end both those types of measurements become critical. Just to reiterate one point I don't remember if Stephanie made today, infectious titer is not product potency. It is an activity type of measurement, but it is not potency, per se, of your product. So you do need to distinguish that in your own minds about what were thinking about when we are talking about particle to infectious titer ratio.

There are the issues of uncertainty in the imprecision of the assays, that is particularly an infectious titer assay, and also the fact that most of the

infectious titer assays that are out there frequently underestimate probably the number of infectious particles because they did not account for the slow diffusion of a particle in solution as a particle is getting near a cell to have that infectious event occur. So, they overestimate the number of particles that might actually have the opportunity to get to a cell, and you have to incorporate that into your calculation of how your raw data gets transformed into an infectious titer.

RCA is not standardized at all. It is a bioassay with one or two cell lines. Detection is by a variety of methods and it is a semi-quantitative assay. It is a yes or no assay right now for most people. The results, in terms of quantitation, are only based on the amount of sample you put into the assay to analyze. Quantitative data would really make the regulators job easier, and it would make our own jobs easier in understanding our own products.

No one is doing the assessments the same way. There's no standard amount or volume of production lot to be tested although there is a recommendation and the guidance for you should have less than a certain amount and

a certain amount of vector. How you get to measuring, that is not described at all in the guidance for adenoviral vectors. There's no standard means to quantify the RCA amount. The standard would assist us in doing that, and there's no standard way right now to report the results, although I think the agency is trying to lead everyone to report the results in the particular standard way. The standard would allow the field, the community to compare, qualify and validate our methods so that the units will actually mean the same thing to everyone across the field. They don't mean the same thing now and that is really the big bugaboo.

The outcome of this October 5th meeting was that the community really wanted an endorsed rapid development of a well-characterized standard, and that the primary standard should be a wild-type adenovirus type 5, and that we do want to develop a secondary standard that will be replication defective. We have not so far spent much time or effort on the secondary standard issue. Hopefully, this afternoon we will begin a discussion of that, but at the moment, the prime thing is to get this standard done and available to the community.

Although FDA is leading the process, the working group is working as a group to make this happen. I want to reiterate a couple of things that came out of, specifically, the October 5th meeting, which will be important for this afternoon's discussions. At the October 5th meeting, the consensus was that particle and infectious unit numbers or information would be assigned to the standard, and for particle concentration, that we would try to use an orthogonal approach to get the best number, because OD 260 reading that is commonly used is based upon a publication with some theoretical calculations, but it's not--how representative that really is a little hard to say.

We thought, from the October 5th meeting, that an orthogonal approach would get us to a number that everybody could really hang their hat on and was scientifically justified. The working group this afternoon will finalize how were going to try to do this. Coming out of that, we expect then to be able to say that here is the extinction coefficient we recommend that you use, if you're going to use the OD 260 method done by a particular SOP. That way these things will relate back to each other, because the OD

260 method with a lysing agent is still one of the most commonly used methods for determining particle number.

Our goal is to get this primary standard available by the end of 2001. That is quite an ambitious goal, but I think it is doable, and so far we are actually still on track, believe it or not. The working group is responsible for identifying the process, figuring out how we're going to do it, and then selecting the groups to deal with the different facets of it; the manufacture, the characterization phase and the distribution phase.

The group currently has representation from quite a few different institutions; including the FDA, NIBSC, ATCC, USP, and obviously the Williamsburg Bioprocessing Foundation. Currently, we have five academic groups, five contract manufacturers, three testing companies 14 Pharma or biotech companies and two suppliers. So, you can see it is across the community. It is not just in one area.

We also have regulators in Europe who are paying close attention and who have asked to receive all the information. They are also keeping up to speed on what is going on here. The group, I do want to point out, is a little fluid, so people may drop out as commitments change.

An institution may decide that they still want to have a representative or another institution may decide to join the group. It is a large group. We are 30-odd people right now, and I hope the group doesn't grow to be 100, but clearly we're not closed in that sense.

Everything we're doing is really meant to be a transparent process in the sense that whether you're part of the working group or not, you have the ability to put input into the process. Being a member of the working group or not being a member of the working group does not mean that you have no way of influencing what's going on. Plus, not being part of the working group does not mean you cannot do one of these functions. The working group is just trying to move the process along and make sure it happens, and that it happens in the timeline that we agreed to, that we feel is really critical to achieve.

The group started out by establishing the list of activities, establishing the criteria upon which selection will be made for each phase, and then we will make a call-out, really through the website and e-mail at this point, for proposals that meet this criteria. Then the FDA will look these over first and then make some recommendations to

the working group, but the working group will be making the decision that yes, we agree, or maybe we do not agree, but here is how we're going to assign the different functions.

All of our decisions, the minutes from our meetings have been posted so far and will continue to be posted on websites. Wilbio.com is the Williamsburg Bioprocessing Foundation's website. All information is posted there. We have gotten an endorsement from ASGT, who is helping us publicize information in their journal. OBA and RAC are also helping publicize information. There are venues that people have to get hold of the information if for some reason they're having trouble accessing that particular site.

So where do things stand right now? We did have a meeting of this working group or the people who signed up in October, in November, and we really made sure everybody was clear on the mission, agreed upon it, and talked about how the group would function and started defining these activities. Just to give you an overview of the activities, we divided it up such that as many--we wanted to make it as flexible a process and as discrete steps as possible, so that as many wanted to participate by

potentially have the opportunity to do so, and so that the burden of doing it would not fall on any one particular group, because there is no funding for this effort. This is strictly a volunteer effort, where it is being done for the greater community good.

If your organization wants to participate and you're having trouble justifying what you should do, it really is for the greater good. There are no dollars. There is no funding for this. It is strictly volunteerism. So obviously some of these things are more costly than others, and that is why they are all listed as donations. Donate characterized cell back files. Donate a source material that meets certain criteria. Donate production of the virus bank. Donate production of a purified, formulated balk that would go to a facility that will donate their vialing services, which the standard will be frozen back and prepared for storage prior to the major characterization phase, where we determine the infectivity and particle number units to be assigned.

There is some characterization that will occur on the balk, as Stephanie mentioned in her talk and Steve mentioned in terms of what we're looking at, but the

particle units and infectious titer units will be assigned based on the frozen standard. There will be other characterization that we want to do on the frozen material, which we will be talking about more this afternoon and then some kind of on-going stability phase that will occur.

I was not going to talk in detail about the requirements, though I can, if either during the panel, go back to these slides, but so far we've talked about very specific things meant to be--allow some flexibility, but meet, sort of, a notion that we have that we really want something that is characterized, that is well understood how it got to where it ends up being, and that it, sort of, meets a certain high expectation that we have. The community really wants this to not be just something that was done in a laboratory and thrown together and then it's out there.

We really want this to be something that could continue. That is why we have a virus bank, so that you can go back to that material and make additional lots in standard, and that it meets certain requirements that we feel are necessary to have as a group. The group has been

explicit about that they really want this to be very well characterized.

One thing that is not part of our mission--our mission is not currently to standardize any of the methods. That is why we're looking at this orthogonal approach for particle number and although we would like to come out of that with an extinction coefficient that might be more useful to the community, we are not trying to standardized any of those methods, nor standardize infectious titer methods or RCA methods. That's really not part of the mission at this time.

We are also not meant to endorse any specific method for producing the standard. Any specific cell culture, viral culture, purification formulation or analytical method, none of that is meant as an endorsement. However, any of those procedures that are used to create the standard will be part of the information about the standard. It is important to understand we all need to know how the standard got to where it was, but the working group is not saying that the only way you can make an adenovirus is this way, the only way you can formulate an

adenovirus is this way, but there will be information about how that ended up being done.

The next steps are--actually, this afternoon's working group meeting, which is very much a working meeting to really focus on, to finalize the call for proposals for the initial manufacturing portions of creating the standard. Those bids should be able to go out after today's meeting, which will also decide, finally, what the deadline is for receiving those bids, so we can move that process along.

The other thing that we will be doing this afternoon is really focusing on the characterization phase and coming to agreement on exactly how we're going to handle that. That is probably going to be the majority of the discussion this afternoon. Those are really the immediate next steps.

I just want to come back to why we are doing this, and really I think the community feels very strongly that comparability between quantities is the key issue for all of us, and that certainly the standard will allow us to do that. Whether you use the standard to characterize an internal standard you may have that relates to your

product, which is probably how most people ultimately will do this, and validate your own methodologies, it will at least allow the field to compare, qualify and validate our methods and then make these comparisons that relate to safety and efficacy.

I think that is the end. I think that is the end for now. I would certainly be happy to entertain questions, either about the specifics of where we are on some of the calls for proposals or any other aspect of the working group.

FLOOR QUESTION: As we all know, actually there is a standard vector and infectious ratio issue as well as wild, type but, there are also major differences in the vector used for gene therapy. I understand there is the confusion on how you classify first generation, second generation, and third their generation. For each one, at least there is modification that makes the vector safer and also, there are cell lines with additional modifications to provide specific production procedures. For example, vector--present toxic genes in their cell line to actually helping to generate higher titer viruses. I was wondering

how we can standardize or put those factors into the consideration of this process.

DR. HUTCHINS: Well, in a way we are not, because we're focusing on three key methods where quantification need to be improved, in terms of having comparable units. We're focusing on particle number, which unless you change the coat proteins in the vector particle, it is irrelevant, anything else that you have done, because the intact particle will be the same, basically, by most measurements.

Infectious titer is the one where--that is really going to be influenced by what type of vector your product candidate looks like. All the infectious titer unit will allow you to do is come up with units, then, that you report back that are meaningful, relative to this standard. You may have an assay--the specifics of your assay require a very specific cell line that complements your product because it's deficient in all these various genetic components. That is fine. In the end, the units you report from that assay have to be meaningful relative to the units of this standard. That is the purpose of the standard.

The purpose of the standard is not say that my stuff is, suddenly, no longer as infectious, it's just that the units will mean something. I think that is the issue that we're trying to get across, similarly for RCA as an impurity. I think during the panel maybe Stephanie and Steve might want to come back to this issue about how they see this for the different types of vectors, but, certainly, we're focusing on units, coming up with units that would be comparable.

FLOOR QUESTION: I wanted to get you to elaborate a little bit more on your slide about what is not part of the mission, where you're saying that you're not standardizing methods for characterization of the standard or the way you are making it. It seems to me that your production process, the purity of this material, and in the way that you--I mean, the assays you're using to verify that are directly related to the utility of the standard. I was wondering, could you give me the rationale behind that particular attitude?

DR. HUTCHINS: Although products are defined by the process, the standard, certainly, is not similar to products, because it is a wild-type material. We decided

that it has to be a purified material, and we want it to be a certain quality and characterized material, but we, actually, do not think for the most part, in terms of how it's going to be utilized in those three key assays that is a critical factor, in the sense that it was grown 293 cells or A 549 cells--or that it was purified by a three-column process or a one-column process, as long as it gets to a certain criteria. We have been developing criteria and maybe where I should go back here for a moment is to some of the criteria.

FLOOR QUESTION: I guess what I'm focusing in on is it seems to me for this vector to be really useful to everybody, you're going to need to have it extremely pure and have a high integrity of all the intrinsic properties of adenovirus.

DR. HUTCHINS: That is correct.

FLOOR QUESTION: Perhaps I'm getting into details that are best discussed in the working group--

DR. HUTCHINS: Or details that, in fact, have already been discussed in the working group.

FLOOR QUESTION: Particularly the purification process--cells are not terribly relevant, as you say. You

are also going to collect data. You mentioned using this orthogonal approach with several different laboratories to try to characterize the thing and define actually what your standard is, and that is going to be dependent on the state of validation of your assays.

DR. HUTCHINS: That is correct.

FLOOR QUESTION: So, I'm kind of surprised just by that statement.

DR. HUTCHINS: Well, the working group may decide in the future that--the gene therapy may decide that it's time to have some kind of standard method to do something. For instance, for retroviruses, through the same kind of collaborative process, FDA ended up with the guidance that was issued, I think, in November, finalized guidance on RCR testing. And, while that does not say you have to do this, put this, it does talk about the amount of vector you should be testing for RCR, at what stage in manufacturing, and it talks about how you would validate the assay and the approaches that you need to take in using the standard to come up with something that has a meaning.

The working group might, down the line, take it as a charge for itself, but a charge that is not part of

its current mission, to address that same type of issue or to apply that to one of these types of methods. That is not part of the current mission. The current mission is to get a primary standard out by--as soon as we can, and then to look at getting a secondary standard out that is replication division.

Let me point out to you a few features that maybe address this point a little bit. For the purified formulated balk, we have looked at how much we want to have, we're trying to leave it open, but we have expectations. We want people to describe what they're going to be doing and a description of the methods and the specifications, and some information about the methods. You can't just say, "We're making up a method to do blah, blah, blah." That is probably a proposal we're not going to look highly upon.

It has to talk about their experience in doing these things and really, even for the formulation, we spent a lot of time discussing this at the last working group. We don't want it to be limited to something, but we want data to be supplied, if you are the group proposing to do this, about what buffer you're finally going to put this in

and why you have chosen that and some data to support that. We're asking for data to support why you want to do what aspect of the proposal you're proposing to do, so that it's not just your reputation. There has to be something to support your proposal.

Similarly, we're doing that even at the virus bank stage or at the--the initial source material has to meet some particular information. And, the cell bank files--we're asking very specific things. Characterization phase--we have not finalized what the criteria should be. That is actually one of the things we're talking about this afternoon. We started discussing that. We have some ideas about where we want to go. But, standardizing methodology--no, that's currently not part of the mission. I hope that answers your question a little bit.

FLOOR QUESTION: Good morning. I would like to bring a different issue. First of all, I would like to just thank and congratulate everybody participating in this effort. I think it was really badly needed. Everyone working with adenoviruses are going to realize they're going to end up with RCA in their hands. It happens. It happens through homologous recombination. One way to solve

that is to move to a different cell line or a different system in which your host and the vector will not have homology and therefore will not generate RCAs.

When you do that and you find out your system works in that sense, you end up with a new cell line, which if you follow the guidelines, which are very clear, both by the FDA, European authorities, Japanese, do everything by the book and yet you find hurdles along the way, such as prions. This is an issue that has come up and is not being discussed here today.

I was hoping to be able to get some clarification as to how the authorities think about the issue of prions, and if it applies to cell lines that were generated, for instance, in Europe, but what you are able to characterize and follow the pedigree and follow the use or not use of serum and finally adapt it to serum-free medium; what are the issues? What is the problem? How can we solve this? What kink of test can we apply? Should we be working on tests?

DR. HUTCHINS: That is not really a mission of the working group at this point in time and I have to turn

that back over to the agency, in terms of how they are thinking.

FLOOR QUESTION: It is related to the biosafety--

DR. HUTCHINS: Yes, but is not related to the current mission of the working group and what you're talking about is something broader relating to production technology. Although that's a very valid question to be raising, it is really not part of what we're dealing with today, this afternoon or--

FLOOR QUESTION: I would just like to seed that into the minds of people, and if anyone from the FDA here--

DR. HUTCHINS: Are you asking that maybe there could be some type of gene therapy community group that you think would be valuable to have that type of discussion with the agency? Is that what you're actually asking?

FLOOR QUESTION: I believe so. I think this is very timely. I understand from today's discussion that RCA continues to be a safety concern. The only way to move away from them is to have a different vector and packaging cell line.

DR. HUTCHINS: Actually I would disagree with you.

DR. SIMEK: I realize what you would like is an answer. The agency is very concerned with the issue of TSC and BSC, and the agency, our division in particular, is in the process of trying to--we're having discussions and trying to develop some sort of internal policy, but we can deal with other cell lines that might decrease the level of RCA. We are not, today, in the position to answer any questions about this. I can assure you we're working on that very diligently and that soon we will have something available that we can make available to the public and to manufacturers about this issue.

FLOOR QUESTION: When you say soon, do you mean within this year?

DR. SIMEK: I just cannot tell you anything.

FLOOR QUESTION: I don't I want to continue the discussion on this point, but I think it is important to keep in mind that scientific interpretation of data may be partially influenced by an interest of the people in developing a certain product. So, I will caution people to say I do not believe there is a concern here or there, because that may be framed within a particular frame of mind. Let's put it that way. I would do like this

discussion to be open in the working group to make sure people are aware of what type of development occurs, so we can interpret comments and the data.

DR. HUTCHINS: However, I will be squelching a lot of references to vaccine this afternoon. So while you can make comments, please realize that it is the gene therapy community.

FLOOR QUESTION: I wasn't referring to the vaccine at all. I wasn't referring to the cell substrate, because clearly if there is an investment in developing a product in 293, then that can frame a type of interpretation of data.

DR. HUTCHINS: Just remember the primary standard wild-type Ad 5. The group decided upon that for various specific set of reasons, but the cell substrate in that sense, in an RCA sense, is irrelevant. In any case, the standard will be useful for methodology to better understand RCA, but it's wild-type adenovirus. That is the primary standard. Cell substrate issues, per se, are not really relevant to this afternoon's discussion. Any other questions?

MR. CARSON: We are a little ahead of schedule. Why don't you stand up? Take about a five-minute break. There is still some coffee left. Please eat the rest of the cookies, because we have already paid for them. We will get the panel set up here and we will start exactly at half past the hour.

[Recess.]

MR. CARSON: Again, the purpose of the panel discussion is to give you another shot at the speakers,.

DR. HUTCHINS: I'm going to hide behind them.

MR. CARSON: To ask any relevant, tangential, otherwise, questions. That's all I had to say. Now, I'm going to turn over the panel, but Stephanie Simek actually has one clarification she'd like to make.

PANEL AND AUDIENCE DISCUSSION

DR. SIMEK: I'd like to make this to for the transcript just so it's clear to everyone. I in no way want to give the audience the impression that the FDA's making the decisions on selecting the working--selecting who manufactures, distributes and characterizes the standard. Our role is to serve as guidance and make

recommendations. The working group itself will make the final decision on selection and all the steps in the manufacture process. I want to make that clear.

FLOOR QUESTION: I have a couple of simple questions. Both Steve and Stephanie used comparability in your--and I want to make sure we understand. We're assuming that comparability as you presented here is not as we define it in the protein therapeutics business, and in terms of having some facilitation of manufacturing changes and that sort of thing.

DR. SIMEK: No, I am sorry. I use the word loosely. I mean, for everyone, for manufacturers and investigators, as well as ours, just to be able to better compare between different clinical trials and products, but we're not talking about comparability assays.

FLOOR QUESTION: That word has alluded definition in the industry.

DR. SIMEK: I'm sorry.

FLOOR QUESTION: The other is a relatively simple question that may go into this afternoon's discussion, as well. Is the goal here to develop an adenovirus standard or an adenovirus 5 standard?

DR. HUTCHINS: It is actually to develop an adenovirus standard and your company is one of the ones that has sort of a hybrid-type of vector system where you have Ad 2 and Ad 5 elements, primarily Ad 2 elements. It is just that Ad 5 is much more commonly used, and so the consensus--we don't want to develop ten standards, right now. We wanted one to get the ball rolling. The consensus from the October meeting was that, really, we will pick one. We will pick the replication competence thing. We will go with the wild-type. Who can--for what we want to use it for, it should really work for most everybody in terms of those key assays.

FLOOR QUESTION: It was reminiscent for me of the interferon situation where we had five or six interferons on the market, and trying to decide which want to use.

DR. HUTCHINS: I think the flavors of interferon is not quite the best analogy, because they have quite different biological activities. Ad 2 and Ad 5 are very similar in terms of their infectivity profiles and other types of issues. Functionally, they are not really different. Interferon alpha, beta, gamma really are.

FLOOR QUESTION: I have a couple of questions for you guys. One, I will play the doubting Thomas, and I would like to hear a little bit more about the RCR standard, being in the vaccine area. To my knowledge there have been international reference standards proposed for almost every virus that is licensed, but I can't think of one, but maybe polio that actually exists. Despite trying to coordinate labs and assays and so forth, it just has not happened. What is the difference in this effort that we are actually going to get something that is quality and accepted by regulators for adenovirus?

DR. SIMEK: First of all, this is not going to be a WHO standard so we're not looking at it in that sense. This standard--you have to look at it, or at least that we look at it as in perspective of gene transfer studies. We have to start there. We cannot start any bigger than that. For gene transfer studies, this will, if we can decide on testing and the agency accepts the qualification of this, will be used as a standard for gene therapy trials. That means that, under the best circumstances, CBER will accept it for that. Will it be accepted for other things? We just cannot answer that and that wasn't our goal.

FLOOR QUESTION: So you're really just looking at this in order to establish connectivity between clinical trials, and not necessarily use this as a gold standard to look at CMC type.

DR. SIMEK: No, actually, if this standard is produced within our recommendations, I mean, we would accept this, most likely, for use as a reference standard for Phase III and clinical trials of gene therapy products. Okay. It will also be used by investigators. We can go out everywhere for gene therapy products. Anything above that, we are not in the position to make that decision. That is not what the role of our working group is.

DR. HUTCHINS: Getting back to your point about viral standards in general, NIBSC is re-examining their thinking about that issue and they are having a meeting at the end of March to look at--reevaluate where they are, where they might want to be for viruses. They are looking at both gene therapy and, I think, more broadly than that as well. Tony Meager[ph.] will be participating by telephone this afternoon, but I do not think anybody's here in person from the NIBSC. I can give you more information about that, John, if you are interested.

There may be, sort of from the world community point of view, a more broad look at whether we need to have some other types of standards. Those are not specifically gene therapy standards, just viruses that are available that mean something.

FLOOR QUESTION: My second question was, just on the notion that several industry concerns have started making noises about having adenoviruses as well-characterized biotechnology products. I would like to hear from the FDA people as to just how far they think that can be taken.

DR. SIMEK: Do you mean specified products?

FLOOR QUESTION: Right.

DR. SIMEK: No, the agency is not considering, at present, adenoviral vector as being a specified product.

FLOOR QUESTION: Could you elaborate on what would have to happen to enable that?

DR. SIMEK: No, I cannot at the moment. I am sorry. But that is not even a consideration at this time.

FLOOR QUESTION: Sort of following up on that last question, would the standard that we are proposing to make, also act as a reference standard for things like

process residuals, such as residual host cell DNA or BSA-tween, any other process residuals?

DR. HUTCHINS: I can tell you from the working group point of view, it is not currently what we factored into it. So, no, we're really looking at three assays where the standard would be useful. Because we are allowing aspects of it to be very open, in terms of we're not saying it has to be made by a certain process or certain cell line, I don't think it would be relevant as a standard in those other areas. I think you would probably agree on that.

DR. BAUER: I think we agree completely on that, the infectious titer, the particles and the RCA are really our interest, right now. I do not see how you would tie your kinds of concerns together with that at the present time.

FLOOR QUESTION: Since we're focusing on the RCA, and we also had a discussion about what defines RCA, I think the definition is very clear, replication competent adenovirus. The virus has to be able to replicate in the absence of the gene being supplemented. So, in the cell line the virus has to be able to replicate. For example,

in the first generation adenovirus, it is very clear that the virus has to carry E1 gene. There is a PCR procedure that is easy to detect, if the E1 gene is within the viral backbone or still in the cells.

I think the value for the standardized--the infectivity, I say, will be relatively difficult, because how much virus can you add in the cells, even the replicate defective virus, will cause the cyto-toxicity in the cells. I think it should be easier to actually create the general standard and to define what is RCA, and then the next issue will be what is the level of RCA that potentially can cause problems. The difference between vector and RCAs--RCAs actually produce viral proteins, which we know is very strong--to those actually in nonimmune-compromised animals or human, those viruses would be eliminated.

So I agree, if you have too much, the replication competent virus will cause a problem. I think that is probably what we should focus on. First, we can define. It is not different--to say RCA is different in different situations. RCA can be easily defined. For example, there are vectors with E4 deletions. If the E4 region is in the viral backbone, is replication competent virus. If it is

not in the backbone, is not replication competent virus. There are ways to define it, if it is a supplement gene in the backbone or not. PCR is the best, easiest way to use.

I do not have stock in the company, but I think it would be the best way to do it. Once we define what is RCA, then we can define what is a tolerable level in the stocks.

Also, I think it was an the issue as to what the cell line uses. We should not care what the cell line uses. You can use the cell line and have no replication potential. That is better, as long as it can produce a vector that reaches the low-level RCA, then that is allowable. I think it would be clear for everybody to follow, otherwise is kind of very confusing.

DR. BAUER: I just want to make one comment about your PCR comments. I think designing PCR studies to correlate between a piece of DNA being present and the ability to replicate is going to be tricky. I am not saying it is impossible, but you can have fragments of DNA in a preparation that would come positive on PCR, but not necessarily on infectivity assays. Having confidence that your infectivity assays are working and have met some

performance with a standard, you can then start to do the kinds of correlation studies you're talking about. I think that is another potentially useful aspect of having a standard with a titer that people agree on.

DR. HUTCHINS: You have to use the microphone.

FLOOR QUESTION: I think your point is well explained, but I think there is potential to generate pseudo-positive in the PCR production, but it would be on the safer side. I would rather generate a false positive than a false negative, that is number one. Secondly, there are ways to avoid just a small piece of DNA to be amplified and also, for example, with the increased sensitivity, a lot of technology being developed to detect low titers of HIV infection, there is nested PCR that can be detected in very low numbers. I think if we can, at least, set a goal, what is a plausible standard to see if there are any problems associated with this type of standard. If there's no other problems, then we will have a standard that can be applied to everybody.

DR. SIMEK: I mean, you're right. If you use the PCR assay, in our opinion, it is the worst-case scenario. So that gives us an idea of the absolute maximum on RCA.

But, you have to understand that when you start getting away from your defective vectors and you start talking about your replication selective vectors, which we're not going to do with this really today, and you do PCR assays, the number you get might be quite high, and it might be alarming where in reality that doesn't really represent infectivity. So we're all for the worst-case scenario, because it gives us a better idea, but we'd have to discuss it, I guess.

DR. HUTCHINS: I think the only way you can distinguish those for selectively replicating vectors is, in fact, to look at the molecules that are present and not focus on their functionality, because we don't have assays that can discriminate those very readily. So, that is a separate topic, but the fact is that molecular definition could actually assist in understanding the differences on a broader basis for Ad-vectors.

DR. SIMEK: Right.

FLOOR QUESTION: I was going to just comment on a somewhat contrasting situation, but I think it touches on comments made by the other doctors. It was stated that there were no standard viruses, but for--I think the one

that was picked out, polio, while perhaps there is--others can comment, perhaps he does not feel there is a standard. There is a standard method for evaluating the safety of polio and that is a neurovirulence test. In the performance of those tests, there are standards that are used in the corporate world and, certainly, within CBER.

That assay is so important that it is done by both sponsors and the agency. There, the assay is critical. I think that we cannot forget that. I think it speaks to the feelings that we don't need to be concerned about a particular assay when we're talking about the use of a standard. I just wanted to point that out. Even agency experts who've led the development of molecular methods for polio virus will not--would blanch at the thought of using a molecular method for the evaluation of polio with regard to its potential neurovirulence. That is the maverick test.

I think we cannot forget the importance of having, at least in this case, a safety test that we're going to rely on, and not just that we're going to be concerned about the standard that we're going to use in

that assay, just some thoughts on maybe a slightly contrasting situation.

FLOOR QUESTION: I was wondering if you have any concerns in terms of the list of donations that need to be provided in producing a standard. Obviously, you have a set of criteria. You want a high-quality standard. Are you concerned at all that this list is not going to be met, that people are not going to come forward with these donations, because in order to get the best quality of standards, what you're asking people to give to you is a huge investment in work that's already been done, and how do you see this coming together?

DR. HUTCHINS: I know informally--I know that for every phase so far, we're covered by at least one potential donor. That is informally, because we have not actually sent out the proposals and asked people--the call for proposals and asked people to send the proposals in yet. It is an issue, but I think we're going to be okay. The one phase that many of us were concerned was the vialing, and we do have a group who says they will put a proposal in and it should be able to do it and meet everything that we've talked about so far in our criteria. That was the

one area that people were most concerned about getting a donation contribution from, because a lot of people will not allow an Ad 5 wild-type--they might do that in a production area, but they will not allow that in a vialing area. That was one area, but I think we are even covered there. It is an issue, but I think we will be okay.

DR. BAUER: At the October 5th meeting, I think a lot of us were very impressed with the spirit of volunteerism and the willingness at that meeting and subsequent meetings to support this. We're all confident that it will go forward, even with the expense and the burden of doing the different steps.

MR. CARSON: Any other comments?

FLOOR QUESTION: You made it clear today that your intent is to develop a gene therapy standard. It would be very helpful for us to understand the criteria that defined the standards as gene therapy standards, and not just the fact that it was sponsored by the CBER division that is dealing with the gene therapy.

DR. SIMEK: What I meant by that was that all I can say at this present moment is that the standard that we are considering will be used for INDs and, hopefully, for

future licensure of products used in gene therapy clinical trials. I mean, I understand you would like this to go across the board, and at present we represent one division and one office in CBER. We do not have the ability to assure you that this is going to be used anywhere else. Certainly, once we can get the standard developed, it is open to the rest of CBER to look at. If it meets other offices qualifications, of course it could be used. It is not restricted, but, again, we cannot make that decision for other offices and will not. We cannot tell you that this will be accepted. We can only say that it will be considered, I am sure, unless everyone would like to use that, but that's not what we're here for.

DR. HUTCHINS: Talk to Bob.

DR. SIMEK: Yes, honestly, we're not, all the offices discuss things with one another. Everyone is aware of this. We have representatives here from CBER. It is not closed, but we do not have the authority to make that decision for them, nor will we.

FLOOR QUESTION: I was wondering if Dr. Anderson could comment on Office of Vaccine.

DR. SIMEK: I don't think it's appropriate to put him in a position like this. That is not what this working group is for and I really don't think that is a good thing to do. I will tell you that afterwards, when we get this together, clearly, we will discuss this with the other offices. I don't think it is appropriate to go in that direction right now.

FLOOR QUESTION: I agree with you, it is not appropriate to put him on the spot, because, obviously, I represent Merck, but I also express my own opinions, so I would not make a public statement that my opinion represents Merck. I do not expect Bob to comment on this.

On the other hand, the comments that were made before on the validity of the standard, in a broad sense, is a big issue. The people that are involved in developing a standard; if there is, perhaps, too narrow of a focus, may not take into consideration other issues that may be helpful in actually developing a standard that is useful across the board. So, perhaps, one way we can address this concern is to have a working group, an effort to actually broaden the participation of the people involved in developing the standards, so that there is also

consideration to the other issues that will be very relevant.

I think it is important to do this before having a standard already established, because as you know, it is much more difficult to change something that is already established.

DR. HUTCHINS: I think the urgency for the gene therapy community may not allow the luxury of time to revisit more specifically some of those--whatever those issues are that the vaccine community might have. As a representative from that community, I--energize, organize your community to do what you need to, and be aware, observe, comment. I do not think we want to go back from where we are right now with the working group.

We really want to move ahead with due speed, which means releasing the first set of proposals as soon as possible, the first call for proposals as soon as possible. We do not have the luxury of time, I think, to delay the development of the standard in the adenoviral gene therapy community more than it is going to take us to get there. It is going to take us to the end of this year to get there.

I think a lot of us feel very strongly that, and Stephanie and Steve can comment on the agency's view on that, but the gene therapy community feels very strongly about that. Unfortunately, I am sorry, but you have your own community within which to work, energize and collaborate with the agency on. We will hear your comments. We will try to deal with that, but we can't start the process over at this point.

DR. SIMEK: That is correct. Right now, we're dealing with a need by the public, by OBA, and by the gene therapy community, and that is what this effort is for. I know it may seem that it is restricted and, clearly, there will be communications, also, among other offices in CBER. I cannot stress enough, that is not what we're here for today.

DR. BAUER: Just to second Beth's comment, it provides a model of how this kind of collaboration and interaction can take place, which I hope is used much more widely in the future for a variety of products.

MR. CARSON: I need to make a couple of, I think, important announcements, like getting you to the right place for the afternoon. Someone pointed out to me, on

your diagram--right behind your agenda is a diagram of the campus. Even though we say Building 29B--have you look at this recently?

DR. BAUER: The B is not on there. It is a wing of Building 29A.

MR. CARSON: It is actually 29A, isn't it?

DR. BAUER: Yes.

MR. CARSON: Do you see that little circle on top of 29B? That is where the front entrance is to the building. Do you see where I mean? You will come in that door and sign in and get a badge. Security has been notified to have adequate badges available, but, again, it is on top of what is called 29A. It is certainly very walkable from here. We will start at 1:00. Again, you will have to be issued a badge.

Participation--again, seating is limited. What we're doing is we have conference rooms A and B. They are very close to the guard desk when you come in. They are right there, the conference rooms A and B. You can be directed by the guard. He is at the registration desk. If we need to, we do have an overflow room, which is conference room C. Again, the Fire Marshall will only

allow us to put so many people in the room. Over and beyond that, we have set up the speakerphone so that people can hear what is going on if that is needed. We will fill conference room A and B on a first-come, first-serve basis.

DR. HUTCHINS: No, the working group members and priority.

MR. CARSON: Excuse me, working group members, we have to give priority to, yes.

DR. BAUER: They will be around the central table.

MR. CARSON: I misspoke. We will, certainly, give seating at the table to the existing working group members. There will be a priority of sorts, a pecking order of sorts. Again, lunch locations: Second Floor, Building 10; Building 31, which is out of your way, if you want to do that; then, of course, there is a cafeteria just down the hall. Thank you very much.

[Luncheon recess at 12:01 p.m.]

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