those checklists were indeed filled out for every patient registered. So we considered that perhaps there seems to be confusion about what we mean by monitoring the conduct of the protocol versus implementing the protocol and getting the procedures in place before the first patient is enrolled.

And that's what I mean about frequent confusion regarding the distinction between those procedures to start the protocol versus adhering to the protocol as written which is something that goes on as patients are being enrolled in regular review.

Next. After we clarified what we were looking for in much more explicit detail, the subsequent series of letters and communications we've received to the INDs indicate that, in fact, there are very few deficiencies in terms of the programs which are described in their ability to actually meet all elements of good clinical practices. The deficiencies that did exist were few, but they included both issues of procedures and description of organizational structure or staffing so that what I will describe to you in the second and sometimes third rounds of communication between the FDA and the IND sponsors, the kinds of things that people still seem to have trouble making sure that their monitoring program has

in place.

Next slide.

DR. SALOMON: Patricia, may I interrupt for just a --

DR. KEEGAN: Sure.

DR. SALOMON: I guess what's really bugging me right now is the -- maybe I don't have this right. But what I'm looking at here is that were 20 -- you sent out this letter.

DR. KEEGAN: Yes.

DR. SALOMON: And 26 INDs covering 64 protocols were reviewed.

DR. KEEGAN: No.

DR. SALOMON: And then you sent out a subsequent thing. This is a whole year.

DR. KEEGAN: Yes.

DR. SALOMON: And after a whole year there are still 106 INDs that are active with insufficient information to assess the monitoring program.

DR. KEEGAN: Uh-huh.

DR. SALOMON: And 32 new INDs have been submitted and 16 of them are active with some attempt to address the March 6th letter. I guess when you go back a slide and you say there were very few deficiencies, are we talking about then this small

subset of 26 that you can evaluate because you've still got four times that many that you haven't got information back yet.

DR. KEEGAN: What I'm talking about is yes, on the 26 and in addition some of the 106 were still going through it, but on the second review of the responses which again we haven't collated in full detail, so I couldn't give you the numbers on that. On the second time around, people usually get it, but I can't give you the exact number where we've gone through and ascertained that everything is absolute and complete, other than for the first round, but on the second round we generally have.

DR. SALOMON: Okay, I hated to interrupt you, but just for me to be processing what you're presenting, we're talking about a study that's not complete yet, that you have maybe 25 percent or 30 percent maybe by now, I'm just guessing, close to that and based on that 30 percent, you're giving us some feedback.

DR. KEEGAN: Right.

DR. SALOMON: So all these statements about there aren't that many deficiencies, etcetera is based on this subset of total -- then I can sit back and --

MS. LAWTON: Can I just comment on that though because I understand that you're also providing us feedback on the additional INDs that you've had answers on the second round which -- so it brings it higher than percentage. It's not just the 26 INDs. It's the others in addition to that.

DR. KEEGAN: It's the others, but I can't give you a firm number for that. This is basically in discussions with the staff. Like I said, when we sort of closed it out and put it officially in our database as where the review stands, then I'll have better numbers, but it's in terms of trying to do that. Again, as regards to process, you should recall that the March 6 letter gave sponsors up to three months to respond. The number of responses that we got prior to June was a handful. I'm estimating less than 10. So most people waited until the last second. Many of those people, I should say that there were a number of people who didn't even respond to that, so we had to send out a second letter, basically putting people on notice that if they didn't do something, we would put their INDs on hold. So by the time we had information in to begin our review, it was really the summer of 2000. So it's taken a while to get through the number of INDs and protocols. So I think it's just the

process of getting through this and giving you numbers is the issue and I'm supplementing it by the flavor of the responses on the second round for which I don't have solid numbers.

DR. SALOMON: Just for the record I in no way mean to criticize presenting preliminary data. We do that every week in my lab. I wanted to make sure that I was sitting here listening with the appropriate context.

DR. SIEGEL: Let me put this in context because it's, I think, a little less preliminary than you may think. I hope so because we're talking about thousands of hours of reviewer time to generate it.

The Agency and I'm not talking about just gene therapy or just biologics, but the Agency as a whole has always required that clinical trials be monitored and that there be QA and QC, that there be assurance that there's good clinical practices in following the protocol. That's a sponsor's responsibility and periodically either for cause, but most commonly at the time of licensing, we inspect to ensure that that, in fact, the trial had been adequately monitored, or more importantly we judge the success of the monitoring by ensuring that the documentation do support the fact that the data are of

high quality and that the patient's welfare and rights were appropriately protected.

What we have not done and again, I'm speaking Agency-wide, what is pioneering about this effort is we have not asked sponsors to tell us up front for our review how they go about doing that and have not reviewed those activities, rather we've trusted that they do okay and then post hoc at the end when they come in for licensure, we inspect to ensure that we can trust the data and also again, checking for patient protection.

As many of us, Dr. Zoon and myself sat in discussions with senior officials at NIH and at the Department and in the period of the winter of 1999 and 2000 and looking at some of the things that we had discovered at some of these inspections and some of the concerns that were being read and also the loss or significant loss of public confidence in the ability of medical researchers to protect patient safety and welfare and rights, particularly potentially in the area of gene therapy, we began to look at what could be done to better assess the situation and better determine where the problems were, improve the status of events and also potentially if appropriate, restore public confidence.

So this approach of asking sponsors to describe their monitoring techniques represents, if you will, a pilot effort, something the Agency has not engaged in before to any significant extent. We did require it, but on the other hand, recognizing the novelty of this and the difficulties of responding as well as reviewing to these data, we implemented it with a certain amount of flexibility. We were asking for a lot of data and then we asked for it in a two to three page summary. We weren't highly specific and I suppose aside from the fact that we had good reason to expect something better than we got, we also had good reason to expect that we didn't know exactly what we were asking for and that sponsors didn't know exactly what to provide, simply because of the nature that this was something new and we were -- so what developed was an interactive process to get at what we felt would be the most important information to know and what sponsors and the most important thing for sponsors to do.

Now part of what we discovered is that there were a subset of sponsors just as we discussed somewhat about academic sponsors involved in the manufacture of products this morning that some academic investigators involved in the conduct of

clinical trials, the concept of quality assurance and quality control and independent oversight of their activities was a relatively new concept which isn't to say that they weren't doing good clinical trials or safe trials or protecting patients, but the concept of independent oversight and documentation in some of these same principles which is what we have traditionally looked to for assurances that that happens, was relatively new and so the answers we got back, I'll make a long story short, but the answers we got back to the initial round of questions, as you'll hear more of soon, reflected a broad range of to some extent lack of clarity on our part, but also of just not understanding what the issue was. You know, quality assurance, I thought that was the FDA's job or the IRB's job or something like that. And so we've got that -- if we didn't get back substantive and workable and reassuring responses on people within a couple months of the three months' deadline, there were clinical holds. So there should be no suggestion here that three quarters of the people haven't responded a year later and they're still conducting trials. That's not what's happened. But what has happened is first of all a lot of their responses indicated that they were describing systems that

they've had implemented since receiving the letter or since the headlines in gene therapy, so a lot of this involved implementation of new systems to ensure quality control and a lot of it involved -- well, they respond, but they maybe missed some points we were interested in and we'd get back to them and say we really want to hear more about how you're doing this and so forth.

So to say three quarters is incomplete is true. On the other hand, there's been 100 percent review of these responses and those trials that are on-going are in a position that we're comfortable with where they are.

DR. SALOMON: Okay, without any further discussion -- I appreciate that clarification. We'll get back to that because I have some questions on that and I think Ed had a comment. If you'll accept my apologies then for interrupting, Patricia.

DR. KEEGAN: My concern is just that I hesitate to give numbers where I don't have firm numbers on some of these issues. But at any rate, those areas where we found that again some of the plans on more detailed failed work, I'm sorry -- go back a slide. Go ahead. All right.

This is actually a summary of the

description of the monitoring procedures that were described there. We found that there was a lot of variability across the board that monitoring visits might vary from weekly to annual, that monitoring visits in some instances were not tied to the calendar, but were tied to patient accrual. That was a relatively uncommon situation. More often it was really tied to a calendar. That the proportion of patients' records that were reviewed and verified for accuracy also ranged, and it was variable. It ranged from 10 to 100 percent. Again, in some instances it also varied by the phase of the study or the size of the study.

Next slide. In terms of the concerns that we had where people still needed to doa little bit more work, there were failures. Probably the most frequent was failure to describe actually the individual who was responsible for directing the investigational drug product to make clear whose job that was. Sometimes there was also a failure to put in details about the procedure itself. Failure to describe the procedure for removal of investigators who failed to adhere to the protocol is written in the GCPs.

Next. No procedure described to ensure

that the modifications were reported to the FDA. Again, not that it may not be happening, but that they didn't describe the procedure. No procedure to describe for verification the study information against the source documents or for how they maintain the study records and not providing a procedure to ensure that the safety reports are filed to the IND. This last one is the only one that raised just a little bit of concern in that that was one of the few where it wasn't simply a lack of information, but where there was some -- in some instances some misconceptions on the part of the investigator, that if they filed it to the IRB, the IRB would send it to us. Or if they put it to MedWatch, it would end up in their IND. And in those instances we did make sure that people were contacted and understood that they had it wrong and what they needed to do to correct that immediately.

In terms of the clinical monitoring staff, again, a variety of arrangements that this basically covers the waterfront here. Frequently, particularly if you're a sponsor-investigator, it's a research nurse or team of nurses who report to the investigator. Also, at academic sites and this seems to be a relatively recent phenomenon that many sites

now have a clinical site team that reports to some individual at that study site, for instance, an administrator, and that they perform a service for investigators at that site to do monitoring and auditing, that there's monitoring staff that's employed directly by a commercial industry sponsor, that there are contract research organizations which perform this either for sponsor investigators or commercial sponsors or sometimes it will be a combination of the above, particularly again for the smaller biotech companies or even for larger biotech companies that they will have their own staff and it sometimes also employed the services of a CRO.

Next slide. In terms of training and qualification of the monitoring staff, this seems to be fulfilled primarily by training as a health professional. In some instances commercial sponsors and CROs also have developed their own predominantly on-site separate training programs for the individuals who do monitoring for them.

Next slide. The concerns in the clinical monitoring program that rose to our review that are -- and again, this is a rare instance. I think there's actually a very limited number of sponsors, I want to say or one or two, who transferred monitoring

obligations to the CRO, but failed to maintain a copy, so they weren't able to give us much in the way of a summary of the CRO's procedures for fulfilling the obligations. However, they did verify that they had reviewed those procedures at the time of the contract and felt that they fulfilled all their criteria for monitoring.

And the other which I believe you've heard about before is the fact that there are sponsor investigators directly supervising the monitoring staff which raises concerns about the ability of a monitor to implement corrective action for somebody who is her direct supervisor.

Next slide. In terms of commercial sponsors, again, we found that there's been a problem very limited, but a few commercial sponsors who have acquired other industry-sponsored or academic programs where there wasn't any details about monitoring and they don't really have much information about studies conducted prior to their acquisition of the studies and that raised a whole other set of questions about how much background work they needed to do to investigate particularly older studies.

In terms of the impression, and again, this is for the 200 INDs which we have, at least,

preliminarily looked at and had discussions, most of the sponsors have staff identified to perform monitoring and auditing. There is a variable frequency of monitoring and a variable amount or extent of data verification, how many search records are evaluated, how many patients' records of the proportion to patients in a trial. And again, variable degree of independence between the clinical monitors and the investigators.

The impact of the variations in the conduct and organizational structure of the monitoring programs on adherence to GMPs is not clear from our review. We don't know if it matters, exactly, whether the frequency or certain types of programs make a difference. It is clear where we have specifically asked and received a response that there are a number of sponsors who have augmented and approved their programs in the past two years.

Next slide. With regards to the preclinical and this will be much briefer. There are 135 INDs where the response has been reviewed and deemed to be completely adequate. In 119, the sponsors verified that all safety information had been submitted. For 14, the sponsors actually supplied some additional information and in some instances it

was just publications of previously reported information and in others it was actually new information that we had not seen.

There are two sponsors which have summarized -- it's actually one sponsors with two INDs who summarized additional information, but hasn't provided the raw details and they have been asked and have verified that they will be supplying that shortly.

There are 39 INDs or master files where there's the responses were incomplete and they've been asked to clarify what exactly they meant by their response. The most common was well, it's not applicable to my file and we didn't often know what precisely they meant by that, meaning it's not applicable because I did it or it's not applicable because I don't have any animal studies or what, so we have asked for additional information and there are 16 that remain where I don't have the results of the review yet, where they're under review and I'm not sure if they were adequate or what the actual outcome was.

The majority of sponsors appear to be in compliance with the applicable regulations for submission of the animal safety studies and the only

question raised by reviewers was the ones where sponsors said all our information is contained in our cross reference file and it was sort of a parenthetical by our staff that they were not certain to what extent all IND sponsors are completely aware of everything that's contained in the master file. They certainly don't have any right to be aware of everything and so on occasion our response is we hope that you have that in writing, that you'll be aware and have that in confirmation that all animal studies are being appropriately reported.

Next slide. That's it.

DR. SALOMON: Excellent. Then I'd like to go forward without any more discussion to Dr. Salewski, Chief of the Bioresearch Monitoring Branch who is going to talk about the exact overview of the subset centers that we've done on site. And then we have a series of questions that I think are clearly extraordinarily important to this discussion this afternoon.

MR. SALEWSKI: When I was asked to present to this advisory committee I asked to see the roster of the members and I didn't recognize anybody's name, so I decided a brief overview of the Bioresearch Monitoring Program might be helpful to everybody

involved.

The purpose of the Bioresearch Monitoring Program is to ensure the integrity and quality of the data that's submitted to the Agency in support of a marketing permit. That includes INDs, NDAs, IDEs and ensure that the rights and welfare of the human subjects are protected.

In FDA, each of the five centers has an active Bioresearch Monitoring Program. Currently, it's coordinated by the Office of Enforcement. That will change relatively soon. There's a new office in the Office of the Commissioner called the Office of Human Research Trials where all of the programs under Bioresearch Monitoring will be oversight responsibility and coordination responsibility will be transferred to that office, except for the Good Laboratory Practice Program which will remain in the Office of Regulatory Affairs. And all Bioresearch Monitoring Programs are conducted by field investigators, occasionally accompanied by an expert from the Center when we feel the need for that expertise.

There are four programs associated with the Bioresearch Monitoring Program and as you can see we have oversight of product development from the

animal testing stage through the clinical trials associated with marketing applications.

When do we become involved with biologics? We mostly get involved, most of our work is associated with license applications. On occasion we do get referrals from CBER staff when they have concerns about how a study is being conducted or how they're not getting appropriate responses from sponsors of clinical investigators and after a while they'll come to us and ask us to help them correct the situation. Sometimes other centers, if they find they have a problem with the clinical investigator or an IRB or a sponsor, they'll notify us in case we have any protocols being conducted by those people or research being conducted, so if we have concerns we could also go out and take a look.

Also, recently, I mean in the last two or three years, we've had a real upswing in complaints. We get complaints from sponsors about clinical investigators. We get complaints from IRBs about sponsors and clinical investigators. And I have consumers up there and by consumers I mean participants in the clinical trials or their relatives. They felt they'd been mistreated or not -- didn't get the appropriate test article, so they come

to us those kind of complaints and ask us to resolve any issues. And also we get a lot of complaints from former employees of sponsors and IRBs and clinical investigators who at the time they were working for them thought they were doing the right thing, but once they had to find other employment, they decided it wasn't quite right, so they thought they'd let FDA know.

And then there's the routine surveillance. We haven't really conducted much of that over the years until recently for the gene therapy initiative was our first real routine surveillance try. A typical cycle for a BLA in our center, a Bioresearch Monitoring Representative is part of the committee, the licensing committee. This is just a typical overview of it and the committee member discusses with the medical review officers and the scientific review officers and the statisticians what their concerns are for the trials, what trial sites they think they'd like to go see. We develop an assignment. We send the assignment out to the field. The field will go out and od the investigation. They'll write up an EIR which is an Establishment Inspection Report. They'll send that to my group. We'll evaluate the EIR. We're write the appropriate correspondence and then after we

get all the inspection reports associated with the license application, we'll develop a summary document which will provide to the licensing committee detailing what we found at each of the clinical sites and with the recommendation to either accept the data or reject the data from one or all the sites.

Next. How do we go about selecting the sites? Basically, we'll sit down with the reviewers and see what their concerns are. The goal that we shoot for is that we try to get the sites that have treated at least 50 percent of the patient population. Sometimes we can't do that because there are some trials that are huge like the TPA trial had 60,000 subjects treated at over 500 sites. So we couldn't quite do that. And there are other trials where they've treated maybe 110 subjects at 87 sites. So we don't have the resources to do that. So we'll get together with the statisticians and come up with some kind of scheme to do our inspection with.

But basically, the higher the number of subjects at a site, the more likely we are to go and inspect that site. Also, the geographical distribution plays a part in our selection. If a license application has 10 clinical sites, six of which are in California, we may end up only doing one

or two of the sites in California. Also, we look at the inspectional history of the clinical investigator. We'll not only look at our database, but we'll contact the Center for Drugs and look at their database to see if that person had been inspected before and if he has, what type of inspection, what kind of problems did they find at the site. If it was a violative inspection it's most likely we'll go back and look at that clinical investigator to see if he's changed the way he's conducted trials. If the reviewers note inconsistencies in data such as too many adverse reactions at one clinical site or not enough adverse reactions at one clinical site or if the data is being driven by one clinical site, we'll basically go see those places.

What we do because the field, there's many other things other than wait for a Bioresearch Monitoring Inspection assignment. They also do blood banks. They also do warehouse inspections. So instead of going in there cold, we like to give our investigators some information so that when they go into a clinical site they know what they're looking and they know what they're looking for. We tell them what the product is, how it was developed, who the sponsor is obviously, what patient population this

product is being given in and what the expected outcomes of the trial are. We ask them to look at adverse events, see if the protocol was followed, see if all the subjects met inclusion criteria and not exclusion criteria, see if the blinding was maintained throughout the study. We checked to make sure that the appropriate dose was given at the appropriate time frame and did they meet their end points.

And after they go through all of this, after they perform their inspection, they'll sit down with the clinical investigator and go through with them before they leave which we call a close out, before they close out the inspection they'll sit down with the clinical investigator and discuss with them the findings, this is what we found that you didn't follow your protocol, you included several people who met the exclusion criteria. And they'll discuss it with them and they'll make this part of their report that they send to us. After they leave, they'll write up this EIR. They'll send it to us. We'll classify it. We have basically three classifications, no action indicated, voluntary action indicated, where there are several violations of the regulations, but the violations really didn't affect the data from the study or violate the subjects' rights or welfare.

Then there's official action indicated where it's met a threshold, where the data has been affected by their conduct in the study.

What we do is we will issue correspondence. We have basically two types. One is the untitled letter which goes to the NAI investigations and the voluntary action indicated investigations. We'll write to the clinical investigator or the sponsor and say this is what we found at your site, how do you plan to correct it in the future? And then we have titled letters. One is a warning letter where we say this information, the violations here are affected. What happened at your site? You have 15 days to tell us how you're going to correct this or tell us why we're wrong in our assessment.

Then we have this Notice of Initiation of Disqualification proceedings and the opportunity to explain, commonly called the NINPO. By the way, it took the Agency 14 months to come up with that name and you know it's a good name because nobody likes it.

And this is where a clinical investigator will get this notice once he meets the threshold of deliberately violating the regulations repeatedly. It's "or repeatedly violating the regulations or

submitting fraudulent data to FDA or to the sponsors." We'll initiate that disqualification procedure and of course, they have the opportunity to explain and if we accept their explanation the matter will be dropped. If we don't, we go ahead. We proceed with a Part 16 hearing.

What can we do as far as administrative actions? We can recommend that the data not be accepted to support the application. We can recommend that they refused to file the BLA or put the IND on clinical hold or terminate the IND. In compliance, we can't actually do those. We make recommendations because it's the scientific review staff that makes the determination of whether to place someone on clinical hold or terminate the IND.

However, as far as disqualification goes, we have the authority to go ahead, go forward with disqualification or the application integrity policy issues. But we do that in conjunction and the support to the medical and scientific staff at our centers. We don't go off on our own and do this. It's a joint decision, it's just that we end up with the work of doing it.

Okay. And now, the gene therapy inspections. After the inspections of -- in

Philadelphia and in Boston, the Center was concerned about the state of gene therapy investigations in the United States. So what we decided to do was take a randomized sample. At the time, there wre 211 active INDs. We -- after consultation with our statisticians we determined that a number, 30, would be appropriate and so we selected 30 INDs in a randomized fashion and we extracted every principal investigator doing a study in each of those INDs. We ended up with 24 sponsors and 70 clinical investigators. So we basically issued 70 assignments to look at how clinical investigations were being done.

The breakdown is here. As you expect, most of them are independent with only six commercial sponsors and as you would expect the commercial sponsors had the most clinical investigators associated with their INDs at 46 and I thought you might be interested in this. We asked the field to do these inspections within 60 days. We didn't quite meet that time frame, but the field spent over 4,000 hours doing these clinical investigations. That meant they spent between three business days and 26 business days in the clinical labs, in the doctor's office, looking at their records with an average of 75 hours and that's equivalent to four and a half -- what we

call full-time equivalents in the Agency.

And what we found was this. Washouts are places where they hadn't started treating subjects yet, so out of these 70 clinical sites, 11 of them were washouts. The classifications are broken down there which we're pleased to see that there are only three really violative inspections and again, just so you know, voluntary actions, we found some regulations of the regulations, but they didn't reach a threshold where we take an action. An official action indicated where there was only three of those, again, where we actually took administrative actions.

And for the commercial sponsors, this is the breakdown of the left most column is the breakdown within those four to six. The overall is within all the gene therapy inspections. So as you can see, there was most of them had some violations of the regulations, but not enough to warrant an action.

The government, of course, we do a better job.

(Laughter.)

Next. And the independent clinical investigators which actually kind of surprised me, they were doing very well. Our inspection, I guess I should clarify. Our inspection just looked at how

they performed their clinical trial. We didn't look at monitoring. We just looked at this is your protocol, did you follow it? Did you do all the appropriate paperwork? Did you notify people of adverse reactions? You kept count of your drugs and your patients? That's all we did. We wanted a snapshot to see what was going on.

And this is a comparison of what we find, in general, as compared to -- with the gene therapy inspections. As you can see, that's fiscal years. Fiscal Year 2000 includes the gene therapy inspections and the one below that is without the gene therapy inspections and you can see that on average, even though these were Phase 1 and Phase 2 studies, the investigators were doing a fairly decent job on following the protocol and taking care of the patients' rights and welfare.

Next slide. And what we found the most common violations that we found and the most popular one was not to follow the protocol. That includes things like enrolling subjects who didn't meet the entrance criteria. Not giving the appropriate does or at the appropriate time. Not doing appropriate lab work, etcetera. And then there was problems with the consent forms and lack of supporting data for the case

report form entries, etcetera. As you can see, and these are basically in line with what we find in our normal course of business. They're no different than anybody else. There's no surprises. The surprise for me was that they were so good, actually and that was pleasant.

I think that's it. Do you have any questions?

DR. SALOMON: Joe, just two quick things. What's a washout?

MR. SALEWSKI: A washout is when they hadn't started treated subjects.

DR. SALOMON: Okay, and then the last thing, I just want to make sure I understood this right. Under GT inspections, a comparison, I think your third to the last slide.

MR. SALEWSKI: Okay.

DR. SALOMON: So GT was gene therapy and 2000 total was just all of your bio actions?

MR. SALEWSKI: Yes.

DR. SALOMON: In the year 2000?

MR. SALEWSKI: Yes.

DR. SALOMON: Good. I understand. Okay, I think we better delve into this before -- there are a series of three questions that I've been given to

generate discussion on what is clearly a very important issue.

Before I bring up the questions which take the group's discussion in specific directions that I'm going to try and hold you to, is there anyone who feels that they just have to make a brief, underline the word brief, comment overall? I mean I've certainly taken the liberty and I won't deny anyone else on the committee to do that. But if -- so I know that -- do you want to --

MS. LAWTON: I actually had one question for the presenter and that was I was interested to know with this comparison for the gene therapy trials that were audited compared to the other trials, do you have a feel for the ratio of kind of Phase 1-2 trials that you looked at compared to Phase 3 trials normally?

MR. SALEWSKI: Normally, that comparison was what we usually look at are Phase 3 trials. So these being Phase 1, Phase 2, they turn out very well compared to what we see.

DR. SALOMON: Okay, any other questions? All right. So I'd like to go on record as saying that it is really, the message is reassuring as I hear it based on the data today that after all the publicity

on -- and concerns by the public that the rate of serious violations and conduct done during what's clearly a very rigorous review with hours spent at each center is actually half or less the general violation rate, depending on how you did it. And I think that's pretty remarkable.

MR. SALEWSKI: I just want to add that the Philadelphia sites and the Boston sites weren't included in the gene therapy results. It was totally different.

DR. SALOMON: Right, well, that certainly wouldn't have been random either.

(Laughter.)

In fact, if they had been I think we'd have to start all over with the idea of how you randomize this which we've let you go on. Okay.

So the questions, the first question is really a critical one and it's going to take a little bit of reading to set the stage for, so forgive me. So the regulations acknowledge that the sponsor of an IND may also be the clinical investigator. In that case, they're referred to as a sponsor investigator. The FDA wants us to consider, however, that it's difficult to understand how a sponsor investigator is capable of performing certain required tasks and it's

evident that the experience recently in gene therapy has had everybody take a much harder look at this.

Specifically, the regulations impose that an IND sponsor or sponsor investigator who discovers that an investigator is not complying with the signed agreement, that the general investigational plan or the requirements of this part are applicable to parts, blah, blah, blah -- that this investigator now should promptly secure compliance or discontinue shipments of the investigational new drug to that investigator and end the investigator's participation in the investigation.

Well, the obvious point here is is that if you're the investigator, it's kind of a discussion in the mirror.

(Laughter.)

And that's obviously an issue of major concern.

Secondly, a sponsor shall select a monitor qualified by training and experience to monitor the progress of the investigation. Now here we realize that in practice that has meant that that monitor is typically a research nurse or a research technician employed fully by the investigators or sponsor/investigator and we've already begun -- Dr.

O'Fallon, for example, pointed out to us the obvious problem with that. These people work for us. They want to please us and in fact, something that -- no, it was Joe mentioned I thought was really, really critical and that was that an increasing number of complaints are from research nurses or monitors who had left the employ of the investigators and now are complaining to the FDA. I think that was definitely something worth repeating.

So please discuss the relative merits of various approaches to the oversight monitoring. So given the potential concerns with monitoring programs in which the monitors directly report to the sponsor/ investigator, I think that's what I've just articulated, should these be discouraged?

If such a program is utilized, we should discuss what, if any, additional elements or safeguards could be employed to ensure adequate oversight and minimize conflicts of interest issues, etcetera.

There's a second part of this, but let's start with that.

DR. SAUSVILLE: So I think this is really a proverbial fox and henhouse sort of question and I think that one approach that might bear some thinking

is that the institutions, either universities or hospitals or what have you which are the sites at which these studies are conducted, might be in the position of serving that bridging or intermediary role. My own view is that to have a research nurse work for the investigator who's studying the entity that the research nurse is monitoring and if that's the closed loop is that that needs to be strongly discouraged, if not actually made -- I hesitate to use the word illegal, that's not our role, but I mean at least in some way made not a normative procedure. I think that the institution which is at one level another type of sponsor of the research should be charged with putting in place a monitoring system for the studies that it undertakes by its investigators, that the cost of that is going to be figured into the indirect costs, either for grants or for other funding arrangements and that the monitoring service, in essence, report to the institution. The institution is then in the position of serving as an ultimate watchdog who would hopefully balance the fox and henhouse relationship.

I don't know that that actually has been put into practice, but that strikes me as one model in which we might get around some of these issues.

DR. SALOMON: That was very nicely stated, so let me just make sure that -- so one possible reaction here that Ed has articulated very nicely is that we just advise the FDA that this is not an acceptable relationship in the future and then -- and that's one thing we should just decide. That doesn't necessarily mean then what it is we should suggest in its place, but we should parse this out that one comment is this is not an acceptable thing.

Now the second thing, also well articulated, is that we should allow the institution to use indirect funds and other resources within the institution to provide that service for investigators within that institution. I see those as two different things, both very important for us to discuss.

Dick?

DR. CHAMPLIN: Just one thing, the obvious thing here. The research nurse job actually isn't monitoring and the research nurse's fundamental job is to be conducting the research, generally screening patients, eligibility, etcetera, collecting data, making sure that the samples are collected and that the credence given according to the protocol.

DR. SALOMON: That's monitoring.

DR. CHAMPLIN: Well, that is actually

doing the study and collecting the data. Now monitoring is a second function that is -- it is the oversight function that that role is being done correctly. So I think it's a misinterpretation to say that the research nurse shouldn't actually work for the investigator. There should be a second layer where someone else who is not primarily involved in the protocol is, in fact, monitoring and I don't disagree with the concept that it should be an institutional function because the institution, of course, does take responsibility for the conduct of research activities carried on within its jurisdiction.

DR. SALOMON: Okay, so that's fair. What Dick's clarifying is it's not that there's something wrong with the research nurse. There should be research nurses, but as long as they're identified with actually the conduct and perhaps supervision of materials flowing around, that's all a good function, but the monitor. There has to be a position now that we refer to as a monitor which actually is an important point here.

DR. CHAMPLIN: A fundamental --

DR. SALOMON: That person can't work for the sponsor investigator.

DR. CHAMPLIN: A fundamental principal of quality assurance is you don't inspect yourself or monitor yourself, that there has to be an independent entity and that serves that function, so the people conducting the trial shouldn't be monitoring themselves, but some other individual within the organization should have that function.

DR. SALOMON: That's good. That's a refinement.

DR. PATTERSON: You actually started to talk about the issue that I wanted to bring up. I think it would be helpful if the committee came to a common understanding of what is meant by independence. Are we talking about independence from a reporting relationship? Independence of financial ties? And harkening back, actually, there's a good analogy I think from Mary Malarkey's presentation this morning, the independence of the QC, the testing unit, from the production unit and the QC unit has -- although it may be employed by the sponsor, it has an authority to override in some instances their decision may trump. And I think trying to figure out in terms of clinical trial oversight what those relationships are or are not. Even in the situation that Ed described, one could argue that there may be some institutional

conflicts of interest and ultimately the people reporting to the institution are employed by them. So I'm having some difficulty understanding what it means by independence.

DR. SAUSVILLE: I mean I guess my view about that is that the unit that does monitoring might work for and actually obviously might be employed to a certain extent either -- certainly at least in a contractual sense by the university or by the institution. But I think that the nature of the relationship should be that they are empowered to make their decisions quite independently from the decision making structure that runs the clinical trial and now how one exactly sets that up I guess would obviously bear some thought, but the general principle would harken exactly to what you said. This needs to be viewed almost as an Inspector General or some type of function that is quite independent from the actual operation of the trial.

DR. SALOMON: The problem here though is what follows and that is what -- as Amy points out, what is independent? So an institution, how independent is an institution of its investigators? Now an institution will often hold the patent on the product that the institutional investigator is

testing, so there already there's a -- it's very common these days and that's a major thing. They may even hold stock in the company that the investigator started to run to do these trials and we have examples of that right now.

DR. SAUSVILLE: But that's exactly why, I think that they would be vested in, as it were, getting this right. Because I think that if the monitoring agent were actually independent in the sense that I mean in the limit case they were actually a company that was hired for this purpose. And at one level they're going to get paid whether or not there's a patent ultimately resulting in a product or not. I mean the nature of their relationship is that they are contracted for it.

DR. SALOMON: Right, but one of the recent cases, I believe the facts are correct, at least as I know them from the newspapers is that one of the CROs that was contracted had a stock position, an ownership position in the company.

DR. SAUSVILLE: That clearly then fits into what was brought up before. That's the -- that type of CRO should be intrinsically disqualified from this role.

DR. SALOMON: So how do you generate a CRO

in a university or in an institution, a research institution that can do that?

DR. CHAMPLIN: For example, the University of Pennsylvania has a vested interest to be sure that they don't have regularities in future gene therapy or other clinical research studies. The institution's interest is to remain in business as a clinical research center and it is clearly in their best interest to avoid these kind of events, so that they have a natural interest, to be sure that the clinical research is done appropriately, far exceeding any gains that they have from any individual product being successful or not, so I think that there's much more confidence there at least in my mind than perhaps a small biotech company in monitoring their own clinical trial where they have a much greater financial interest in its success or failure.

DR. SIEGEL: It's worth nothing that although closely related, there is a distinction to be made and I think Amy is right. The issue of what independence plays is very complex, but there is a distinction to be made between the independence, vis-a-vis functional independence and reporting responsibility versus the issue of financial conflict of interest. They're both very important. It's worth

knowing that in the long history of drug development that clinical trials are monitored by the sponsors which are usually pharmaceutical companies and which have a tremendous financial interest in that trial and I think for the most part, but not always that financial interest points toward their ensuring that they get the best, highest quality data and the highest quality trial and good patient protection, but not always, but -- and that -- but what differed from some of the cases we're talking about, well, the levels of financial conflict of interest differ, but another thing that does differ is this issue that those monitors are not working for or with the investigator and the FDA actually has had to tighten up its regulations in this area, but the sponsor has an obligation and is expected to dismiss the -- to act independently and to dismiss the investigator when he's not acting well or to correct those actions or dismiss them as it says in our regulation.

So conceivably, a university such as you're suggesting Dr. Sausville, there may be some financial interest. I imagine there's always some level of financial interest, sometimes more if they own stock in the company, but it's not -- but on the other hand it might well be very different if you have

study nurse monitors who are reporting to and hired by and working for the dean's office than if you had study nurse monitors who are reporting to working for and hired by the principal investigator that are actually monitoring what the investigator does. And as to your question, just one quick comment, you did ask has this been done, is this being done. We're seeing a growing number of institutions, particularly those institutions that either by OHRP or FDA, or the press, have had some bad publicity about their clinical trials, but a growing number of institutions building clinical trial oversight programs, we've got report a number of them are occurring in gene therapy and in your handouts, there are some concerns about are they intensive enough, trained enough and so forth. We think it's an interesting direction to look in. We're all in agreement with I think the original sound advice, the first thing this committee said, you can't very well monitor what you're doing yourself.

I should say one more thing to put this in context. All of these issues are being broadly discussed throughout the country, academia, throughout the department, throughout the agency. There's new policy under development. It's a bigger question than gene therapy, but it has -- a lot of the questions

arose from gene therapy and I say that as a matter of context because on the one hand, this committee and it's advice isn't going to directly lead to a decisive decision, but on the other hand, I think we recognize very importantly that decisions that we make in this area and I and others in the room are quite involved in the committees that will be making decisions in this area, are -- can only be made with really a lot with feedback from the patient and scientific communities. We can come up with all sorts of rules about what universities and researchers can do and I assure you from past experience that we're quite capable of coming up with roles that don't work. And so -- we really are interested in this discussion.

DR. SALOMON: So, so far what I think we've already -- just the way the discussion goes, then unless someone ants to stop here, let me just capture one thought that's clear, that we are advising you that the sponsor should not employ the monitor, the investigator sponsor should not work -- well, that's actually interesting. The monitor shouldn't work for the investigators if there's a sponsor and let's say six institutions under that sponsor, nor should a sponsor/investigator at a single institution, either that an academic or biotech, in either case a

monitor should never work for the investigator or the investigator/sponsor. I think we've all said that. So that's good. We've got that settled.

Then the discussion is going toward who then is far enough away or independent enough to equal -- noble enough to take on the responsibilities of monitoring it, right, and trying to be practical here.

MS. MEYERS: It's supposed to be the IRB. And the IRB's responsibility is not just to approve protocols, but to monitor the conduct of the research.

DR. SAUSVILLE: That's not correct. I mean -- right. IRBs certainly receive reports about adverse events. They judge protocol consents and are very active in human protection aspect, but IRBS, at least in the places that I have been have not involved themselves with the shall we say the technical management, how the clinical trial is being conducted. That's just not their role.

MS. MEYERS: Then they're not obeying the common rule.

DR. SIEGEL: IRBs are charged with monitoring the progress of a trial.

MS. MEYERS: It's HHS.

DR. SIEGEL: I think there's a broad range of interpretations as to what that means. What we're

talking about here and the problem -- part of the problem here is the use of the word monitoring. Because we talk about trials being monitored by data safety monitoring boards or monitoring committees and there's been -- who often, you know, in most cases are in no position to know whether the data they're looking at are exactly the same as what's in the patient's chart or whether the -- for a consent form to sign. They're monitoring, but they're not doing site monitoring. It's an unfortunate duality of the use of the terms. IRBs are responsible for monitoring either because of interpretation or because of staffing. Most IRBs practice that by at least once a year, reviewing the safety reports and adverse events. Most IRBs do not, but they certainly are authorized to. I doubt many at all are staffed to and I'm not even sure that -- they are one of the options, but to actually do what we're talking about, going out and actually looking at what's going on.

DR. SALOMON: I think what we have to realize here is the reality. The reality is that over the last several years, because of the concerns that have been raised, there's just been an explosion of awareness, followed by a near explosion of requirements. And there's no IRB that I know of

that's in any type of position to do this. They'll look at the trial initially. They'll look at the consensus initially. They do not have people that go out and monitor 25 percent of my consents. They will get my -- if I have a series of adverse events, they get reported immediately.

MS. MEYERS: But if you don't report your adverse event, they don't know about it, do they.

DR. SALOMON: That's right.

MS. MEYERS: That's why they have to do the monitoring.

DR. SALOMON: That's why what IRBS now are demanding.

MS. MEYERS: They don't have the money to do it and if HHS understands this, they would put the extra money in the grant funds to --

DR. SALOMON: We're getting there Abbey. What we're saying is that the conventional IRB set up in reality is not set up to do this. That's all we're saying. We're not saying that an IRB or an arm of the IRB that we now might name a monitoring group or an institutional data safety monitoring board for trials isn't appropriate. I think that's where the group is going actually or is trying to get us there.

MS. MEYERS: But it would be appropriate

if the funds were there.

MS. LAWTON: But surely one alternative that we're talking about is if the IRBs are able to see that there's an independent monitor assigned to a study that would give them that competence in the same way as we're talking about.

DR. SALOMON: That's right and that monitor would report to the IRB and that's a very appropriate -- the IRB then would be linked integrally with the whole system.

MS. MEYERS: But when we say independent that again gets back to the thing what happens when the institution owns the company or the stock in the company or a patent on the product?

DR. SIEGEL: Of course, the IRB also is an arm of the institution so it's no more independent than an institutional monitoring group that isn't part of the IRB.

MS. LAWTON: Well, Greg Koski is suggesting that IRBs should not be from an institution, but they should be regional.

DR. NOGUCHI: Dan, I would like to just make one correction. Actually, for this area, other than the product requirements, these are not new requirements. There's not an explosion on new

requirements. There is a vast understanding that there are a lot of requirements that a lot of people didn't realize were there.

(Laughter.)

DR. SALOMON: The correction is accepted.

DR. NOGUCHI: As part of the government, we will add requirements when necessary, but what amounts to what we're talking about is not new ones.

MS. MEYERS: They've been there since 1960.

DR. SAUSVILLE: But that illustrates the education and outreach function that was alluded to this morning. I mean the idea that many -- to me, the statistics were certainly encouraging, as you say, that things weren't worse than they were. The other way of looking at this is 50 percent of the trials had a problem.

DR. CHAMPLIN: My institution has actually such a body, an opposite protocol research that is linked with the IRB and they have, in fact, taken on the job of monitoring INDs that don't have another sponsor in terms of an outside pharmaceutical company or what have you. And in our past experience we found, in fact, the most egregious errors did occur in

the unmonitored single investigator type of projects where there was no one supervising that activity. And clearly this type of approach gives a second look to the conduct of all studies and it's certainly been positive and I think that that model is probably the most reasonable one.

There is realistically no way you can get beyond the institution and have some outside entity now monitoring things without really getting into a very complex logistics that's probably not at all realistic. And I think that as long as there's conflict of interest observation within an institution, those people monitoring and the IRB have no vested interest in the product or the company that's being monitored, I don't really view that there's a problem there. I really don't see any large institutions looking to push something inappropriately for their own financial gain.

DR. SALOMON: Okay, so let's take what's Dick saying and explore this a little bit because it still is how much distance do we have to go that stays reasonable, it can be done practically and yet is done properly. Now Abbey mentioned something that's very interesting, the new head of -- is it OBA? OHRP, right.

The new head, he came out to La Jolla and we met with him and then he gave a talk and in his talk he specifically mentioned something Abbey raised and that was he is suggesting that there be professional paid regional IRBs so that in our area where we have Scripps, UCSD and Salk, for example, and a couple other smaller programs, that we would all have one IRB and that could fulfill this sort of -- just as a counterpoint, there is some discussion going on and I don't think that we necessarily need to settle that, but I think that the committee has spoken pretty clearly here that it can't be someone who works directly for the investigator and/or directly linked back to the sponsor and it could be done -- right now most of us feel it could be reasonably be done in the institution. That's good, you disagree. That it could be done within the institution if there was a data safety monitoring board study monitoring group that answered to the traditional IRB.

Now if someone doesn't agree with that, tell me.

MS. LAWTON: So if I can comment on that you said that the monitor cannot be directly linked with the sponsor and I disagree with that because as long as it's not an investigator/sponsor IND, clearly

the sponsor --

DR. SALOMON: I didn't mean to imply that, right. If the sponsor is a company and they have six investigators and hire a sponsor at the company -- a monitor at the company to go around and see -- that's okay.

MS. LAWTON: Maybe if I can also just comment, based on -- we had discussions this morning about quality control of operations and clearly the reporting structure and the independence of that quality control group on the operations side, this is exactly the same issue for clinical and I would say that you can set up, just like all of the drug companies, biotech companies have had to do, you should be able to set that up in an institution as long as you have the right processes and accountability, etcetera for that to work, but it's how that's done. But there is a model there for it to work.

DR. SALOMON: Okay.

DR. SIEGEL: Well, yes. Part of that appears to be the most problem working -- whether it's sponsor investigator or not is -- when you're talking about working is having a reporting system where the monitoring is to someone in the company independent of

the investigator, but unfortunately one of the areas we've run into problems where the investigator is, in fact, the CEO or the principal stockholder of the company and he's investigating his own product and then it is probably pretty hard for somebody within that company to have the level of independence needed.

DR. SALOMON: Jay, that's an interesting question. If I'm -- the word "sponsor" how is that defined? If I'm the CEO of the company and the investigator, is that a sponsor investigator? A lot of times I'm not the CEO, right? The cute thing is I'm on the scientific board and I tell everyone I don't get any money from the company which is, of course, baloney, but that's how we play it.

DR. SIEGEL: It's probably fair to say that most of the pertinent FDA regulations were written at a time when some of the sorts of arrangements, product development and research were not fully considered and so that's why you would read in the regulation that you're responsible for monitoring your own activities and taking actions against yourself if you don't do them well. Doesn't sort of make a lot of sense in that context. But it was really written with a view to other contexts.

Technically, the sponsor who signs as the

sponsor when they file with the FDA and takes on, therefore, the requirements under regulations and guidance and responsibilities of the sponsor, but frankly in some sense that's almost a non-answer. That is the true answer but we can see the same trial with the same monitoring or what appears to be identical trials monitored where the sponsor is the National Cancer Institute, the Director of the National Cancer Institute, the lab chief in the National Cancer Institute or the principal investigator, but they may well have the same oversight mechanisms and the same thing in business. You could see out of the same group where the sponsor might be the university, an institute within a university, the head of that institute. So in some sense, although we talk about it as the sponsor investigator, more to the point is what Pat was getting at was really what the structures and where the true responsibility lies and that's where we're trying to grow our understanding of is figuring out how to address this.

DR. SALOMON: So trying to grapple with what you were saying and what Jay is saying, in the spirit of the discussion, we don't want a monitor who works for any broad sense of that term, works for the

investigator. And therefore, if the investigator is a stakeholder in the company which is the sponsor, that is also a violation.

MS. LAWTON: Isn't there two separate issues here? On the one hand we're talking financial involvement and I think that's one way you could look at how independent do they need to be because for most of us in industry now, there's the guidance on financial disclosure of investigators and we have standard procedures on how we would check that and how we'd make a decision on using investigators. So that would be one thing. But then the other one is the example that you gave, Jay, where you have all of the different levels, the investigator, the institution, etcetera, all reporting into the same place, not necessarily the financial issue of the investigator themselves.

DR. SALOMON: Richard, do you want to make a comment?

DR. MULLIGAN: Yes, I thought maybe if we kept it to the industry issue, it actually may be more helpful. I think it's getting more complicated with -- the industry has a history and I think it might be helpful to analyze. They have a monitoring system. What are the strengths and weaknesses of that system

and what is the perceived level of independence of that monitoring system. I think the answer is it's complicated and if you looked at it from the academic point of view you'd say this sort of relationship would be unacceptable, but if you looked at it from an industry point of view, this is the standard by which monitoring occurs. And that being the case, you're really talking about almost simply an organizational distinction between the two. It's not really who works for who or whatever, but it's an organization, a safety board or monitoring board. It's almost a title. I think at the end of the day as far as you're going to get from the point of view of truly conceptually what's independent. I'd like someone to comment on the industry standard, maybe Jay, how you look at that because I think you've really got to resolve the industry standard before you go to academic.

DR. SALOMON: But Richard, can I make a comment. To me, the problem with this analogy to the industry standard, maybe I don't have it quite right, but what I'm listening is, see, in industry the monitors are paid for, work for, work within the industry within the business, right? Drug Company XYZ has a monitoring group. The critical thing though is

that they are not working for the person doing the study in the clinic, the investigator.

To me, the problem here that we've been dealing with isn't a problem with the sponsor having the monitors work for them, it's the problem of the investigator having the monitors work for them.

DR. MULLIGAN: I'm not sure that I would agree with that, but I think that the issues of independence and separateness are comparable issues, however you want to look at it. That is, the monitors are within the company. They have all the interest in seeing things move ahead.

I still agree with what you say, but I think that at the end of the day in the academic context, all you really are going to end up being able to do is to have a separate organization and name, a name, a body and I think the issue of who they report to, obviously they should report directly to the principal investigator, but they're going to work for the IRB or they're going to work for the Dean's Office or something. I don't think that that distinction is going to be all that keen.

DR. SALOMON: That was fine and the weakness that got brought up that I was trying to address in exception was the situation in which we

said no, we don't need a separate institutional group because the sponsor hired the monitor. But in the case in which the investigator in the academic institution has a relationship with the company, i.e., on their scientific advisory board, the inventor, the starting scientist, whatever, then in that case, the fact that the monitor was hired by the sponsor could be perceived by the public as getting around our recommendations that there be an independent --

DR. MULLIGAN: I agree. I think there probably then is a consensus that if you don't have the monitor hired by the investigator, if you have it institutionally, however that would be, that's clear what we want to have, right?

DR. SAUSVILLE: I actually would like to pursue the thought -- I think there is two different sorts of model, at least two, implicit in this, in that when you look at the industrial model where the company that's conducting even the early phase trials is going to be the company that ultimately hopes to file a BLA. There, it's in the company's interest to have a very rigorous review and reporting on its investigators because ultimately as we just heard there's going to be an inspection process that they're going to have to run as a gauntlet.

In contrast, that what we call academic investigator or the investigator/sponsor, however we want to call this, it's very unusual for that incident. I don't think it's ever happened that those individuals then would actually go for the BLA. The initial start off is generally designed to hand off at some level these initial observations to somebody else. It's a big company, small company, some other company. And that's where, I think, there really is a difference because at one level the responsibility at that point is going to be out of their hands. And so what we're talking about is these very early Phase 1 and Phase 2 endeavors of ensuring that the investigator to monitoring relationship on every level doesn't compromise obviously safety, but also produces a coherent body of data that then is actually, if there's value to it, able to be moved to an actual production orientation. So I do think there are a couple of different levels, as it were, which investigators related to so-called sponsors in this process. And it is unique to gene therapy, different than what we call drug role.

DR. SALOMON: Dick?

DR. CHAMPLIN: I don't know how uniform this now is around the country, but most institutions

have evolved conflict of interest policies that would preclude principal investigators on a protocol of having an equity or large-scale interest in the sponsoring company and that that clearly is a healthy thing in terms of that potential conflict of interest in that often in the Phase 1 phase is that had indicated there is no company and at that point, you might perceive the investigator having potential conflicts, but at least once a company is involved, I think that that policy of precluding equity and interest by the investigator is a prudent one.

DR. SAUSVILLE: You have evolved. I mean obviously it's been a reactive process and I think part of the reason we're here, actually, is the events that those changes have evolved, as you say.

DR. SALOMON: Abbey.

MS. MEYERS: There was a two-day conference on conflict of interest last summer. It was co-sponsored by FDA and NIH and Secretary Shalala was very, very interested in what it said. But basically I think that everybody agreed with that conclusion, that if somebody, an investigator has an equity interest in a product or a patent, that investigator should not be involved in the clinical trials because it would have the appearance of a

possible bias in the data.

DR. SIEGEL: There was such a conference and Greg Koski is leading a departmental group that is following up on that.

The American Society for Gene Therapy issued, I'm not sure you'd call it a policy, I guess it's a policy, but it's not enforceable in any real sense, but saying you shouldn't do this. The FDA has regulations. They're more focused on assuring data quality and so they focused really on Phase III clinical trials and they don't outlaw such agreements, but what they do is indicate that all such agreements have to be reported in detail to the FDA and that we can toss out the data on that basis, so at the time of a license application. So for those efficacy trials, they probably have had a chilling effect on using investigators with financial conflicts.

I'm not sure though, in the type of discovery phases of research that we're talking about, Dr. Champlin, I'm not sure that there's that much consistency across academic centers. I think there are, while there are some that have been those sorts of relationships, there are others that, in fact, as best I can tell, encourage their investigators to have cooperative agreements with industry and at least so

rumors go. And I'm not sure there is yet a consensus on this issue in the academic community.

DR. CHAMPLIN: I think certainly in the Phase 1, as Ed indicated, the goal is to establish preliminary data that would then justify an outside corporation from licensing a developing technology. So in the context of generating that preliminary data, obviously, the investigator, the inventor has an incentive to make that product as successful.

But I don't see the institution at that point having a major bias that they're going to support in any way anything other than the highest quality research and so having the oversight at the level of the Dean's Office or the IRB, Office of Protocol Research or what have you on an institutional level, I don't see as any major conflict, and I can see as the most practical way to deal with this issue.

DR. SALOMON: Michael.

DR. O'FALLON: I think we've always had a situation where highly successful and therefore influential investigators, whether they had connections with industries, they had a lot more influence than the institution than normal RO1 kinds of guys and so we can't solve that problem. The problem is a personal problem.

I think we have to make a suggestion that some administrative, some process through the administration of the institution perhaps through the IRB which is already in existence, clearly would have to be enhanced. I agree, our IRB is absolutely swamped and all of the people are volunteers, quote unquote.

I think we're starting to micromanage the situation here.

DR. SALOMON: And again, I think that's now -- we don't have to solve all these issues.

DR. NOGUCHI: You're right. You don't want to solve them all, but I bring everybody back to the basic finding that is really driving us here. Although we've discussed about what we did since the University of Pennsylvania incident, what that clearly indicated is that the regulations that the FDA has is, in most part good, but there are situations that need to be dealt with regarding human subject protection, period.

There are models from both the industry side, from the academic side. There are newer models that are being tried. All of them have strengths and all of them have weaknesses, but the fact of the matter is if we agree that many of the innovations in

gene therapy come from the academic situation, what is their piratical approach that we can really take toward that and I think that while we certainly all feel differently about whether one has a better or less advantage, I would just encourage people to try to look to the fact that FDA, in fact, is not making any specific requirements. We have suggested that this may be a useful area for CROs, but as you've noted, CROs are not without their own problems. We've noted that academics have their own set of problems in terms of who reports to who, and yet there are strengths in the situation as well in terms of vigor and energy and other academic freedoms that are useful in the discussion.

Voicing all the advantage and disadvantages is an absolute requirement, what you've been doing, but then the real challenge is going to be everybody's opinion aside, depending on where they come from, that this might be better or worse. For the current situation how can we move ahead?

DR. SALOMON: Okay, so let me stop and try again to summarize what I think the committee is telling you today, with the same idea, step in and tell me you disagree. So I think what we all seem to be agreeing on is that there has to be a monitor for

any study. There has to be no relationship, there can be no direct relationship between that monitor and the investigator or investigators. And that should extend back as far as the sponsor, so a sponsor could hire a monitor and monitor trials by investigators with the exception if an investigator is part of the company that that would be considered a violation of the basic understanding. That the monitoring in an academic institution should be done by a separate group within the institution, acknowledging the limitations that we've discussed in detail that yes, at an institutional level there is a potential conflict of interest with institutional holding of patents, etcetera, but that the nobility of the institution is great enough vis-a-vis the monitoring obligations, particularly with federal oversight, RAC and FDA that it's acceptable and pragmatic, and that that organization should report to the IRB or be the IRB in some new iteration of what an RIB is. But I think frankly, to get people's heads around in academia, you're better off talking about it as a separate organization because if you try and say the IRB can do it everyone is going to get hysterical.

And I think that's pretty much specific. And I should just say from personal experience when we

submitted our grant in March for our retroviral gene therapy program, I set up a DSMB within Scripps that none of whom obviously, they're independent, and we brought in several people from UCSD, so it's not even just institutional. I set up a super DSMB at the City of Hope, so that they were totally non-institutional and they report to the DSMB that reports to the IRB that reports to the three IRBs that reports to the GCRC which has an Executive Advisory Board and an IRB. So I mean -- I think that's what's happening in academia. I think we're getting the message.

MS. LAWTON: If I can just say a couple of things to that. First of all, I still want to come back to a DSMB as separate from what we're talking about currently on monitoring, so I don't think we should make that comparison. It's very different activities that we're talking about here.

I think there is one additional level that you could add on if you wanted to to add some level of kind of comfort around the independence of the monitoring group reporting separately to the institution and under GCPs which is basically what we're talking about here, you also have the need to audit and you could have a totally independent auditing group that that institution also has to

ensure because to monitor the independence, if you like, of their monitoring group. I mean it sounds complicated, but this is basic GCPs that we're talking about. It's just how you set it up for those institutions.

DR. MULLIGAN: I was just going to say that I think you did summarize things very well. I think the CRO that Phil mentioned is something you didn't add, that that could be an alternative approach to it, right?

DR. SALOMON: I agree. A CRO could be done. I guess I'm sort of nervous about saying anything about CROs. I'd hate it to get all the way turned around, that now every academician has to hire a CRO because I can just see that being terrible.

DR. SAUSVILLE: You can just add that to the part of the different --

DR. SALOMON: I agree completely.

DR. SAUSVILLE: It relates to the size of the place. I'm sure, M.D. Anderson is large enough, so to speak, that it could empower some panel to do this. I can imagine smaller places that might actually need to look outside themselves. The general principal is the end result. How you get there, there are different solutions to.

DR. SALOMON: Fair enough, but that CRO could report to your IRB and that could be the institutional link in that case.

DR. SIEGEL: See there, the reporting issue, I'm glad you've commented on that because we have at various times hypothesized that perhaps sponsor/investigators who hired CROs are getting more independent feedback than those who hire their study nurse to do the monitoring, but in fact, if the CRO is reporting back only to the investigator, and in fact, we've seen a problem related to that sort of structure, some rather serious problem, so --

DR. SALOMON: But on that face, you've violated it.

DR. SIEGEL: They could hire a CRO who then could report to somebody who has independent authority such as an IRB.

DR. SALOMON: No. The point here is that again, the CRO, just to keep it simple, Jay, the CRO should not be hired by the investigator. Just like the -- in the concept that we've given you, the monitor should not be hired by or work for the investigator.

DR. SIEGEL: I'm sorry, we're discussing solutions for the sponsor/investigator trial and

there's nobody to hire the CRO but for the sponsor.

DR. SALOMON: No, the IRB can hire the CRO.

DR. SAUSVILLE: I mean the -- I'm going to return to the point that the institution is the platform on which all this is occurring and we've certainly seen the institution does get tarred by the brush of whatever difficulties emerge. So it would seem to me that they should be, the institution should be and I used the word before, empowered, to really step in here and -- I mean it's true that the CRO could be hired by the sponsor/investigator, if you want to use that term, but the reporting goes back to the institution which ultimately gives the investigator the license to proceed.

DR. SIEGEL: Right, so you're not suggesting then, if you're talking about within the institution that -- you're not suggesting a preference as to whether an institution has its own internal employees who are an independent monitoring office or IRB employees or whether they hire a CRO?

DR. SAUSVILLE: How they do it, one can imagine different solutions.

DR. SALOMON: That was the point Richard was making to me and I thought it was well taken. But

the point again should be if I'm the investigator, I don't hire the CRO directly. The CRO should be hired by the IRB or the institutional group, what you want to call it, you know, the monitoring -- institutional monitoring board, the IMB. Great.

(Laughter.)

DR. CHAMPLIN: One plea to try to make this as simple as one can do it. This is an unfunded mandate at the moment, another hurdle that the Phase 1 investigator has to cope with to get an idea off the ground and this is becoming an increasingly onerous task and so to not pile on anything other than trying to empower the institutional IRB or monitoring board I think is probably where we should draw the line today.

DR. SALOMON: Abbey and then Amy.

MS. MEYERS: I just want to make the comment because somebody mentioned FDA's regulations for conflict of interest. I want to say it's the most ridiculous thing I have ever read. I read it about a month ago and it's about a paragraph long and it says that the investigator has to report any kind of financial stake he has in the product or something, so and then the sponsor puts that information into a file and keeps it in his file until the drug or the product

is going through the approval process and then FDA has the right to say we'd like to see that file. So the investigator says I own $100,000 of stock in your company and they put it into a file, you see. The patient never finds out. Nobody knows about it unless after the product is going through the approval process, then you ask about it. It's a ridiculous rule and it should be up front and it should be in the informed consent document.

DR. SALOMON: Actually, all our informed consent documents have that very specifically addressed, Abbey in that you -- item 16 of the Scripps informed consent is the investigator does or does not and if the answer is yes to this question, explain the financial interest.

MS. MEYERS: That's wonderful that your institution says that. I have never seen an informed consent document with a paragraph about that.

DR. SIEGEL: Let me comment on that and I don't want to stand here as a defender or an attacker of the rule in its entirety. I'm sure that each and every one of us could design a different rule that we'd like better.

It's important to understand in viewing that rule that its intent was not, which isn't to say

it shouldn't have been, but its intent was not and it's clearly -- its outcome is not to optimize or ensure protection of patients from financial conflicts of interest. The design of the rule reflected desire to ensure the integrity and quality of the data that support determinations of safety and efficacy for marketing. That's why and -- which isn't to say that the first isn't as important a goal, but however, there's a resource issue, of course, in what the FDA does in terms of conflict of interest and of course, as I know you understand very well, the oversight of patient protection is a complex interaction that involves, of course, IRBs, FDA, NIH, so I will agree with you 100 percent that that rule doesn't do what needs to be done in terms of consent and patient protection, whether that should be a different FDA rule or whether in fact we need something that has a scope well beyond the FDA is, I think, is an important issue that of course, we're not going to discuss here. But I do want to say viewed from the perspective of how can you protect patient rights, yes, you can say that's a ridiculous rule, but the rule is there for a purpose and it does appear to have had some significant roles in achieving that purpose in the sense that even though we don't check until after the

Phase 3 trial is done, we have some rather consistent response from industry that before -- when they learn of these conflicts of interest, before they start the Phase III trial, the vast majority of them will select another investigator or ask for divestiture because they realize that they're placing themselves at great risk if they use that investigator.

MS. MEYERS: Don't you think FDA should know about this in advance, if not after the fact, but in advance?

DR. SALOMON: What I want to do just because of time issues stay on track here. The second of the two parts here, I think we've really pretty much discussed. There is a little bit of a twist and sometimes I'm accused of missing the twist and going on, one of the twists you could put here is should we advise the FDA specifically on what they should do in terms of monitoring the institutional monitoring board, the IMB?

(Laughter.)

Now I don't know whether that twist was there, maybe I've just gotten paranoid over the years, but Dick?

DR. CHAMPLIN: I think for an institution like say the University of Pennsylvania, their role,

their job is to teach students, to generate knowledge and academic activity. Clearly -- and to advance any sort of pharmaceutical or gene therapy product is a very minor consideration for them relative to their reputation for honesty, integrity and their overall value to the community.

And so that I say there's real incentive for an institution to do anything other than do the best possible job of monitoring the quality of their clinical research because that's what their reputation depends upon. And so I see them as the white knights, perhaps, in dealing with this issue in the future.

DR. CHAMPLIN: I think that we've come to an agreement on the committee with what -- the premise of what you're saying is that the institution is noble enough to do this right and that's the premise of the institutional monitoring board. The question, I guess I was just trying to make sure we didn't leave and go on to the next one without making sure you guys didn't want -- is that in a way, that could be a whole lot of stuff could go on and then you could find out you had an incompetent, not an ignoble institutional monitoring board. And so I guess the question about be probably the FDA does want to have some sort of program in practice that does review the institutional

monitoring boards, not every year, but on some sort of a basis.

DR. CHAMPLIN: Actually, the FDA inspected us this week. Spent a week at M.D. Anderson, reviewing our IRB. And we passed, I'm happy to say, but there is a process already in place for just that function.

DR. SALOMON: And if that's considered adequate, then we can move on.

MS. LAWTON: Yes, the only comment that I would have on that is that it's my understanding, yes, we've just been through these inspections because of gene therapy, but there is not the resources at FDA to routinely do those types of auditing. So what we're saying is that we're actually -- we are relying on the institutions to do that, to play that role appropriately. And that's fine if that's what we leave it at, but I don't think we should assume, especially for Phase 1-2 trials, you also heard it's more common to do audits of Phase 3 trials and so it's very unlikely that these institutions will be reviewed and audited for that role that we're now saying they should play.

DR. SIEGEL: Well, that's right, but what I heard Dr. Champlin say and I think it's right, it's

not exactly that the institutions are noble, but that it's, in fact, in their self-interest to do this right. And I think my perspective of the experience of the last couple of years with academic institutions is that while that's clearly true, some have not realized that and some -- which is to say they don't have well functional IRBs or well functional clinical monitoring or oversight and may not realize how much that's in their disinterest until they go through experiences such as five or six major academic medical centers have gone through in the past year or two and I won't name names, but we all know who they are anyhow, at which time and I've talked to a number of university deans and presidents and they all seem to think that, in fact, it is in their interest to do these oversight programs much better, that the harm to prestige and the financial harms as well can be huge.

So I think that a lot of what is needed is also education and discussion and networking and university-sharing experiences and learning from each other and learning from industry and from professional groups and whatever and --

DR. SALOMON: My point, Jay, in follow up to what Dick was saying is if tomorrow we now institute a guidance that institutional monitoring

boards need to be set up at all the institutions around the country, which is kind of what we're advising, something like that or these different alternatives, all I'm trying to say is that if you then think you've got the problem solved, I just question that and there should be some sort of a process then that monitors these institutional monitoring boards. That's all I'm saying.

DR. SIEGEL: No more than having commercial sponsors do the monitoring, solves the problem, there has to be some sort of oversight function.

DR. SALOMON: Right, particularly while it's new.

MS. LAWTON: Sorry, can I just ask a question because one way you say you're checking now is new INDs and annual reports, etcetera, that it's a requirement to document for you how the monitoring will be done and the organizational structure involved in that, so that's one way that you could actually look very easily to see what is in place from these institutions when an IND is filed. And you could go back and do that retrospectively as well, if you needed to.

DR. SIEGEL: Right, indeed.

DR. CHAMPLIN: I was just saying that, thinking that this isn't unlike other things that the FDA does in terms of setting standards and expectations. You don't inspect every blood bank every year, but you set standards that blood banks need to comply to and you would inspect some to ensure that, in fact, those things are being carried out. This would be the same principle. You set standards on what institutional review should be and then institutions are held to that standard when they're occasionally inspected.

DR. SALOMON: Amy and then Abbey.

MS. MEYERS: I have to say that people -- that's the way so many people got HIV and hepatitis. All right, we can't allow this to happen anymore, with gene therapy especially because it's going to go right down the tubes if there are more deaths and more abuses of the system. And we have to do something more carefully because the institutions are not the white knights. The University of Pennsylvania was not a white knight and OHRP has gone in and closed down university after university for all of their clinical trials because the abuses were so bad. So the government has got to step in and it has got to be much stronger than it's ever been in the past.

DR. SALOMON: Amy?

DR. PATTERSON: My comment sounds awfully mundane after that. I was going to perhaps offer a segue to Question 2. I think Dan gave an excellent summary about 15 minutes ago, but I think that Alison Lawton's comment about keeping in mind that there's a clear distinction between a clinical trial monitor and a monitoring board and I think the dialogue is continue to muddy those different roles and responsibilities and I want to put in a plea to the committee when you're answering Question 2 to make sure you're very clear about what you're referring to when you're using the term monitoring because I think it will have a big impact on the utility of your advice to FDA, to distinguish a clinical trial monitor from a DSMB.

DR. SALOMON: Good. Abbey, does anybody want to comment specifically on -- you did, I know, I know.

I think then we can move on to Question 2 which Amy has done a good job of sort of setting the stage for. So the regulations and guidance indicate monitoring should be adequate to ensure data integrity and protection of patients' rights and welfare, but they don't describe either the frequency of monitoring

or the extent, the proportion of the patients enrolled, sampling, for example. In some institutional monitoring programs, a randomly selected sampling of active studies are monitored during the year. It's conceivable that over several years, some studies might never be monitored during the conduct of the trial and only I guess retrospectively.

In those programs where selection of studies for monitoring occurs annually such that a study could accrue patients up to one year before the first monitoring study.

I guess what they're asking us is if we've agreed in the first part that we have to have an institutional monitoring board, how -- what kind of a guideline, what do we expect from that institutional monitoring board which of course is the same thing as if our institutional review board hires a CRO, it's still the CRO is becoming our institutional monitoring board. So if everyone is okay with the concept of an IMB, just so we have the right -- we're all talking about the same thing.

MS. LAWTON: I guess I'm not because now I'm getting confused as to whether you're looking at the IMB as more of a DSMB type or is the IMB overseeing --

DR. SALOMON: IMBC is what we've been talking about all along. It is a monitor. It's not a data safety monitoring board.

MS. LAWTON: Okay.

DR. SALOMON: It's monitoring the trials. It's -- I mean maybe we should define, if you want, exactly what monitoring means. Why don't you start, Alison?

MS. LAWTON: I can go there. I just think maybe we shouldn't use the phrase an IMB because I think that's what's confusing it. I think what we all are in agreement, that we're talking about monitoring and that's separate from an DSMB. Monitoring is going in and checking source verification of the data that's put in the case report forms. We routinely do that 100 percent, source verification, you know, making sure adverse events reported, etcetera, that type of monitoring.

DR. SALOMON: What do you call the group in your company that does that?

MS. LAWTON: That is part of the clinical operations group, that's from the company that would go in. We would have clinical monitors for every single study assigned every site that's involved in that study.

DR. CHAMPLIN: This was usually done for a licensing trial, but not necessarily every trial that's being done with a new product.

MS. LAWTON: I disagree with that very strongly. We monitor every single study regardless of what phase of development.

DR. SAUSVILLE: I just -- maybe this is in the spirit of what was being stated, I mean we've used this term IMB or monitoring board. I actually think that's being more complicated than it has to be. Studies, as was stated, are monitored routinely in a Phase 1 and Phase 2 context, at least by what, for example, studies of NCI sponsors.

And one could imagine that an institution, if the reporting structure, and this gets back to what we said before of the people who are doing the monitoring is separate from the investigator, it doesn't need to be dressed up as a board or anything. I think there are well established ways of source verifying adverse events, reporting, etcetera.

If you feel that we want to layer on this notion that there would be an auditing function or a monitoring function, that's going in, I think, a potentially difficult direction. I think that as long as the general principles are stated, how -- either

companies or institutions solve this, I think, to use the analogy that Dick made before, FDA should set the standards and obviously when these things are called into question in the normal following up of things then if things aren't being done, then it will make itself apparent. And that's where it would stop.

DR. SALOMON: Yes, I have no problem with any of that stuff. I guess I was -- remember, I initially came up with the IMB just to have a word and we congratulated me initially for having quickly -- it just shows you why you can never come quickly with a word because it doesn't work that way.

I like the idea now of the OCM, the Office of Clinical Monitoring.

(Laughter.)

Just kidding. Anyway, the bottom line here is that it's not -- I just wanted to stop us from talking about that being an invisible add-on tomorrow to the IRB, that's all I was trying to get across, but it could be just two or three individuals given some space somewhere who are in charge of monitoring all these programs.

So if we do that, how often should these people be monitoring? Are we talking about weekly, every single patient enrolled, some sort of a

guideline for if it's a 100-patient trial or a 10-patient trial that it would be different?

MS. MEYERS: In my mind, I'm thinking this is going to be the people who go in there and check that the adverse events have been reported to the IRB and to NIH, the RAC, and to FDA, just to make sure that the paperwork is right and that nothing is being kept secret. So I don't think they'd be needed more than twice a year to go in and check and make sure that all those adverse events have been --

DR. SAUSVILLE: To me, it's an accrual-based issue. I mean if you have a very active trial, they're going to have to be working all the time. If you have relatively infrequent accrual they don't have to be doing things all the time. So I mean that's going to be -- generally, there's a percentage type basis, 10 percent, 20 percent of the charts get looked at, that's on the high end. Two percent is on the low end. And people probably sort themselves out somewhere in between.

DR. SALOMON: There's certainly -- there's one more question and there's more discussion that we could have. We're at a point here and particularly because of some issues that need to be done today, cannot be done tomorrow, and particularly with Dr.

Sausville who needs to leave some time around 6, so I'm going to end this discussion here. I think that I've summarized it more than once. I don't think you need to hear me do this again. I'm sure you're all relieved. If we haven't solved everything, I'm willing to at the discretion of my colleagues here bring this up again tomorrow and I'd like to end here for the moment and go on to the end here which is we need to present the CBER intramural research programs and then have -- we need to do that quickly enough to have some time to close the session and have some discussion with Dr. Sausville who chaired that.

DR. SIEGEL: You needn't feel badly about not solving everything, let me just say that. That wasn't the goal, as I indicated. This is an intensive, but on-going and not overnight process of relooking. The whole structures of oversight of clinical research and patient protection and I think the perspectives of this committee are a very important part of that and we appreciate the discussion and I'm sure we'll be talking with you more about it in the future.

DR. SALOMON: This part is still public. It represents the on-going FDA process of site visiting and review of internal research programs and

we have --

DR. SIEGEL: Yes. I am on the agenda, that is, in lieu of Katy Stein, the Director of the Division of Monoclonal Antibodies who is unable to be here today.

And in the interest of time and also because it's really not terribly essential to the process, I'll keep my remarks very brief. As we're entering into the overview of Dr. Marjorie Shapiro, the role of the division director and my role is just to provide a little bit of framework. The Division of Monoclonal Antibodies is one of the three product-oriented divisions in my office, along with Phil's Division of Cell and Gene Therapy and Division of Therapeutic Proteins and then we have a Clinical Trials Division that Karen directs and an Applications Review and Policy Division. And it has as its name would imply oversight of monoclonal antibodies, both for diagnostic and therapeutic use, as well as some closely related products built in monoclonal antibody backgrounds. The science in this field and the technology in this field have been expanding and burgeoning rapidly as many of you know with tremendous advances and the technologies for engineerings these antibodies, designing them, selecting them and

producing them and applying them to various diseases and as such they've represented as many as half of the new products that we've reviewed and this division plays a very important role, both in review of those products and in setting the policies and procedures for that class of products.

Dr. Shapiro works within the Laboratory of Molecular and Developmental Immunology in that division and is one of our investigator-reviewers and I'll leave it at that.

DR. SALOMON: It's my understanding now that we'll get a brief presentation.

DR. SHAPIRO: Good afternoon. I'm going to try to shorten my remarks, so if things don't go as smoothly as they might have, it's in the interest of time.

My interest has been in my lab has been in studying the contribution of individual germ line light changings to the diversity of the antibody repertoire and we've shown that genes that are fully functional in terms of their ability to recombine don't always get used in a pre-immune repertoire. And from this observation, we then went on to start another project because we're beginning to see antibodies derived from new and exciting technologies

in the field such as the humanized mass and fate displaced library* (T6S1-beg.) And they have a vast potential to produce both safer and perhaps more efficacious antibodies. But there may be potential implications that we don't understand about these products, such as they don't undergo the normal selection process that an antibody that's produced in a human or a mouse might go through.

I'm going to briefly skip through this. This is my slide of B cell development which I hope you all are aware of.

Next slide, please. Basically, B cell development hinges on the rearrangement of heavy chain and light chain genes, expression of various forms of the B cell receptor on the cell surface, lead to a variety of processes including allelic exclusion in the pre-B cell, receptor editing, apoptosis and so on as you go on through development.

Next slide, please. This is a picture taken from a paper from Hans Zackov's group which mapped the entire three megabase murine light chain region. There are 141 individual genes which are represented by the mice. Mice here of the same color are within the same light chain family. We've been particularly interested in the three gene family which

is shown there in the oval, the Vk10C family. Two members of this family are seen in a variety of immune responses to both T development and independent antigens in several different kinds of inbred mice, but the Vk10C gene has never been seen in a mature antibody and we've been investigating why.

So the next two slides show the results of our studies. Next slide, please.

The first paper we published on this we showed that the Vk10C is structurally functional and is capable of recombination, that messenger RNA is present in the spleen at 100 to 1,000 fold lower levels than that of the utilized genes Vk10A and B, and an in vitro model using a reporter gene assay, we show that the Vk10C promoter is less efficient in pre-B cells than the Vk10A promoter.

Now we've done some site-directed mutagenesis of the three nucleotides that are different between the A and C promoters and we show that if you change one nucleotide that would be near the transcription initiation site, in the Vk10C gene and change that to the Vk10A nucleotide, we can restore the efficiency.

We then went on and tried some EMSA, electromobility shift assays and those results were

inconclusive, so we're sort of at a dead end for now with this aspects.

Next slide, please. A more recent paper we published we showed that the Vk10C gene is completely accessible to the recombination machinery. It's equally accessible or even more accessible than the Vk10B gene based on a readout of germ line transcripts, that the gene recombines at the same frequency as other family members and the most interesting observation was that as a B cell matures from a pre-B cell through the mature B cell stage in the periphery, you selectively lose productive Vk10C rearrangements.

So the next slide shows some possible reasons for Vk10C expression. The first is that the promoter is inefficient in pre-B cells and because of this you may not get enough light chain protein expressed to pair with heavy chain to put a mature immunoglobulin on the cell surface.

Another possibility is the light chain protein doesn't pair well with heavy chains and again, you wouldn't get immunoglobin expressed on the surface. In both cases, this cell would remain functionally a pre-B cell because it wouldn't have any mature immunoglobulin on the surface, so light chain

would continue to recombine until it made a good light chain of some other gene from some other family and that would be a reason for losing a Vk10C rearrangement.

The third possibility is that Vk10C can pair with heavy chain, but when it gets put on the surface it undergoes a negative selection event. In such a case, again, the immature B cell which is still in the bone marrow, a negative selection event would either lead to apoptosis or again receptor editing where a light chain recombination would continue and again you would lose the light chain gene.

So next slide. At the time of the site visit last October, I had these slides about future directions and I want to spend a little bit of time discussing what we've done with these proposed experiments at that time.

The first experiment, again, is to get back to this inefficiency of the Vk10C promoter. So we thought rather than trying to stick with the in vitro assay and the gel shifts, we would try to do a real time PCR in freshly isolated pre-B cells. And an outline of this experiment is shown on the next slide. All the nucleotides we had used which were specific for the Vk10A, B and C genes in all our other

experiments were not appropriate or the constraints of a real time PCR assay, so we had to go back and develop new primers and probes to do this. We've developed a five prime primer and two different three prime primers which are shown up there on the white that would give us amplicons of 163 and 177 base pairs. And we have three probes each specific for the Vk10A, B and C genes, all contained from within the CDR1 region. And the Vk10C probe differs from A by two nucleotides. It differs from B by 4 and A and B differ from each other by two nucleotides. The other thing we had to do was generate appropriate plasmid to use as controls to work out the conditions. So we now have done all this and we're starting to do the experiments to work out the right PCR cycle conditions and temperatures and everything. So once we work that out we'll go and we'll sort for pre-B cells and do the experiment and hopefully we'll get an informative answer.

Next slide, please. The second future direction was to look, to examine this question of can Vk10C pair with heavy chains. And the way we propose to do this is to put a Vk10CJk1 rearrangement in phage display vector and then clone in PCR of polyclonal heavy chain rearrangements from LPS stimulated spleen

cells. We haven't started this yet. It depends on our ability to make a phage display library and I'll get to that with the second project.

Next slide, please. The third question that we wanted to explore is maybe Vk10C is negatively selected. The experiment design on the bottom, right now it looks like we may not end up doing it based on results we've gotten from the first experiment. The second experiment and I'll discuss the outline of it in a minute, but in the next slide, I'm going to show you results of, we've examined the usage of the Vk10C in autoimmune mice. The reason for doing this is we thought because autoimmune mice are deficient in getting rid of heavy light chain pairs that would be negatively selected in a normal background, perhaps if this was the case we would see increased expression of this gene in mice of autoimmune backgrounds. But as you can see the top 6 mouse strains have the autoimmune background and the last row there is the Vk10 frequency of Vk10C rearrangements in the spleen and you really don't see a significant difference from C57BL/6 and BALB/c mice which have normal backgrounds. So from this experiment it's looking like Vk10C is not negatively selected.

Next slide, please. The second experiment

to address is to look at another kind of recombination called RS recombination. The top line depicts a germ line kappa locus. The greenish box would be the constant region and downstream of that, the black and white box is something called an RS element. It's 10s of KBs downstream. And also in the middle that little black triangle is an isolated heptomer which is part of the recombination signal sequence of antibodies.

Now what can happen is two kinds of recombination here, either a germine V gene, the green gene on the left can recombine through its recombination signal sequence directly with the RS element downstream of Ck in which case you would delete the constant region and any VJ join which would have occurred. And this is a way to inactivate a kappa allele which may have had a nonproductive rearrangement or may be negatively selected for some reason and might prepare the cell to go on and arrangement the lambda locus which usually occurs after kappa rearrangement, but not all the time.

A second kind of recombination would recombine the isolated heptomer in the entron to the RS element downstream of the constant region. Again, this would inactivate this locus, but it would leave a VJ join intact. Both of these kind of

recombinations are seen in 74 percent of lambda positive cells and 12 percent of kappa positive cells. Twenty-five percent of these rearrangements of the Type B which leaves the VJ join intact. And in earlier studies, people have shown that about 47 percent of these VJ joins are in frame which would indicate -- they took that to indicate that these good rearrangements perhaps were eliminated because of the negative selection process.

So we've designed primers and are working out the conditions now in the lab that would amplify specifically Vk10 rearrangements to this RS element and again, we have the primers that we've used in the past that are specific for the three genes. And we would like to ask the question, do we see a higher frequency, a significantly higher frequency of Vk10C in frame or productive rearrangements in this kind of recombination than the others. If it's higher, then this could be taken as evidence of negative selection. If it's not higher, then it would be consistent with our studies in autoimmune mice in that Vk10C is not negatively selected.

Next slide, please. This slide, long term future directions for continuing this study. There are 20 other genes that are functional in terms of

that they don't have obvious mutations that would preclude them from recombining or undergoing splicing or being expressed in any other way, but there have also been no antibodies seen that have used these genes. So we're interested in seeing if the phenomenon of Vk10C is specific to that or if we can find a common reason for why these 20 genes that have been maintained in the repertoire over the years are still available.

And we'd also very much like to get to the level of studying the accessibility of this locus at the level of chromatin. Hopefully that will come in the near future.

So we'll skip the next couple of slides in the interest of time. This is my -- we're going directly to the next project. No, go back one slide, please.

I mentioned before that we have these two new technologies that have vast potential to make antibodies, especially phage display, to make antibodies against antigens that are not good immunogens in vivo. So you can target a lot more things and we see a lot of potential there. But phage display libraries do not undergo any kind of normal selection process. It's totally in vitro. So you

could get heavy light chain pairs that would never come out in a human. So we asked the question is the phage display repertoire normal. And so what we did was we immunized the mouse with tetanus toxoid. We made hybridomas from half the spleen and we made messenger RNA from the other half of the spleen to generate a phage display library. Now as I said, last October, we would like to be as good as regulated industry at making a phage display library, but we're not there yet. We initially had some trouble with the initial vectors that we chose. We have since gotten a new vector. We were having problems with both having high background levels and low efficiencies. When I get back into the lab next week, hopefully we will find out that we've solved those problems and we can generate the library because that is a main goal of ours.

Actually, one of these slides I skipped. Maybe we could just go back one slide, please. What I wanted to say is antibodies are inherently immunogenic. We do have a lot of experience now with licensed products. Our murine products, the whole antibodies, you can see that 55 to greater than 80 percent of patients make an immune response to it. When you remove the constant region, that drops down

to a pretty good level, similar to what you see for chimeric and humanized antibodies. While we don't have a lot of experience with phage display antibodies, there are some hints that they may be more immunogenic than one would have predicted.

Can we go forward two slides, please? So while we haven't made the phage display library yet, we have analyzed our hybridomas and most of our hybridomas bind to the fragment C portion of tetanus toxin and so this summary slide is a little bit more complicated than when I presented it in the fall because we've done some more studies and we're still trying to sort them out. But what we did was we generated 11 fragment C specifics antibodies and two other antibodies, the 18.2.12.6 and the 18.1.7 were generated at CBER in the 1980s and we included those in our analysis. So we grouped them by the VHVL pairs that they express and then we did ELISAs, cross-blocking ELISAs to show that they recognize four unique epitodes on fragment C. We then set up an ELISA to show if these monoclonals could block fragment C from binding gangliocyte which is how tetanus binds to neurons and gets inside cells. And the 18.2.12.6 had been previously shown to enhance binding. In our hands, it did the same. All the

other antibodies blocked binding except for the one on the bottom, 72B9.

Then last summer we started a collaboration with Elaine Neale and Karen Bateman of the National Institute for Child Health. They have a spinal cord neuron assay and so we put our antibodies in our system to see if they could block the activity of tetanus toxin on spinal cord neurons. And everything worked the same as in our GT1B binding ELISA except for the second antibody, 35F7 and the last one, 72B9, where in the spinal cord neuron assay the results were the opposite with what we saw in our GT1B binding ELISA. So we wanted to explore why this happened and we looked at the buffer components and it turns out that the pH has an influence on our GT1B binding ELISA. It didn't change the results of the other antibodies, but for those two that didn't get consistent results, when we started out with our antibodies and a lower pH buffer, then the results of our ELISA were more consistent with the spinal cord neuron assay. And we still don't understand this completely, but that's the data that we have so far.

So next slide, for our future directions, obviously, the phage display library is on the top of our list.

Next slide. We wanted to do in vivo protection assays with our antibodies and affinity measurement. Before we start these those we realize that even though we grew our antibodies in reduced serum medium, bovine serum, bovine IGG has an antitetanus component to it. So we went back and we rederived all our hybridomas in serum-free medium and we're purifying them now. And so we'll get to doing these studies.

But we've spent a lot of time in the last year trying to map the epitopes which we thought would be straight forward and that's also been a problem for us.

Conventional wisdom has it that if your antibodies recognize an antigen on Western Blot, then they recognize linear epitopes. So we contracted with a company -- next slide -- which would map our antibodies on a series of overlapping peptides and these are the profiles. The top two rows and then the panel on the bottom right show the profiles after the isotope controls have been subtracted out. The panel on the bottom left is a gamma 1 control. I didn't have room for the gamma 2 control here. And you can see that we really don't have any good binding. The peaks you see in the middle two panels, all the way on

the right, were also the major peaks in all the antibodies before the back-on was subtracted and if you look -- actually, next slide, please.

Okay, if you look here that peptide falls in this little cavity here in between these two loops, so you wouldn't think that that peptide, that area would be available for binding to antibodies. And indeed, when we had the peptide made, we couldn't show direct binding of the antibodies to that peptide. So this -- these data weren't informative to us other than to tell us perhaps that the conventional wisdom didn't hold true here and perhaps we have confirmational epitopes.

So that we have some other colleagues in the Office of Vaccines that also study tetanus and they have made a series of amino acid substitution mutants and a deleton mutant in this part of fragment C and in the next slide, this is data that we just generated in the last week. I see all my symbols didn't translate.

I didn't name the mutants, didn't specify the mutants because they haven't been published yet, but what we did was we compared their binding relative to wild type fragment C. And in all cases, we didn't have any antibody where it bound fragment C and then

the mutant, it didn't bind at all. It either increased the binding or decreased the binding or what here is shown as the squares. It just stayed the same. So like I said, we just got this data in the last week and we needed to sit down and look at it. Maybe it will be informative, but at a first glance, we may not be able to figure out what the epitopes of these antibodies are. There is one more thing we could try, but we've been trying for a year, so I don't know.

Next slide, please. In our future directions, we have about half a dozen or so antibodies that don't bind fragment C that we want to do similar assays with. We've also rederived these in ceoprime medium and are purifying them. So we'll get those experiments done.

The last slide is our long-term future directions which at this point we haven't begun to even think about yet. And I'd like to acknowledge on the next slide, I have two people in my lab, Sean Fitzsimmons and Kathy Clark who have done all the work, our collaborators at the Institute for Child Health who did the spinal cord neuron assays, Heather Louch and Willie Vanno of OVRR who provided us with mutants.

Thank you.