I can give you several other examples. There certainly is no data that I am aware of that you can just cure patients with early diabetes using drugs like cyclosporine, et cetera. So the argument that this is all an aberration of post-transplant drug immunosuppression, I am not certain I am convinced of that, either.

But I don't know the answer. I am just suggesting that it is another area for research and uncertainty.

DR. SHERWIN: When were those six patients studied in Minnesota with minimal immunosuppression following David's--it wasn't two, as I remember. It was two maybe with on immunosuppression, all of whom had recurrent insulitis and T-cell infiltration of islets after fifteen years or more of post-disease.

DR. HERING: Yes; there are several lines of evidence here. One is the Minnesota pancreas transplants between identical twins. The first patients received no immunosuppression treatment and selective destroyed all beta cells within four weeks.

The first patient had developed diabetes forty years ago. So this is very strong evidence. Then, with immunosuppressive treatment, it was possible to prevent recurrent autoimmune disease in this setting. There is also other evidence, bone-marrow transplantation. So you can definitely transfer the disease.

One bone-marrow donor was, I guess, diagnosed with diabetes maybe ten years ago and then donated bone marrow, and the recipient developed type-1 diabetes soon after bone-marrow transplantation. So that is another piece of evidence to indicate the disease persists.

DR. SALOMON: I mean, the identical-twin data doesn't have anything to do with the argument I was making. But the bone-marrow data suggests that, in one patient, you transferred disease.

DR. BLACK: I just wanted to make one comment to Dr. Ricordi's earlier about the utilization of preclinical data from FDA's perspective. We treat the data on the mechanism of action and clinical rationale lightly understanding that the mechanisms that are present in the animals' etiology of their disease--for instance, the NOD mice, are not necessarily equivalent to the human disease.

We, therefore, feel it is necessary and are also required by the CFR to really only utilize the animal data for phase-I trial design for the point of view of feasibly ruling out what safety problems we can rule out.

Having addressed that issue, that is why I ruled out immunosuppressive toxicity or islet toxicity or infusion-related problems in my talk. But I also wanted to point out, Dr. Salomon, Dr. Kenyon is hoping for a half hour for her presentation.

DR. SALOMON: We had cut that back. I think we will try and stay on time.

DR. BLACK: Okay. Thank you.

DR. AUCHINCLOSS: That was an extraordinary statement. In your view, what you are looking for, is essentially safety data? So you never want to see a mouse study in an IND.

MR. SIEGEL: I think that, in many areas that we regulate of biological therapies, there are immunological issues and species-specificity issues, receptor differences and physiological differences that limit the function of animal models as models for efficacy.

We find that extensive safety data, while sometimes is irrelevant, is sometimes relevant and, therefore, almost always of some value if there is as model in which you can--in some cases, it is impossible to get any useful information from a model. I don't this is the case.

That said, we do look importantly to rationale data. I am thinking back to not long ago when we had a very relevant discussion largely between you and Dan as to how much information one would need for a success of xenotransplantation of hearts or kidneys into non-human primates, say from pigs into non-human primates, before going into humans.

There, of course, the issues of both the

availability of alternative therapies and the availabilities of rescue therapies should the transplant fail are very different from here. But the same issues of how relevant is the model and how much rationale data do you need came up, and, as you will recall from that context, the context of committee and the context of the agency, as I indicated yesterday in talking about these issues, is that where have an anticipated known or unknown risks to a patient such as the risk of immunotherapy or the risks of removing their heart, we will want to see some amount of rationale as appropriate.

As appropriate is put in there to take into account the fact that if there are animal models of very limited relevance, that may be very limited or it may come from other sources, as Dr. Ricordi referred to. There may be therapies for other related diseases in humans and whatever that may provide the appropriate rationale information.

But I would say, for many of our therapies, including this one, if the animal models are not particularly informative, we don't ask too much proof from them. As you will come through our questions, we are going to be asking you specifically how informative are they regarding immunosuppression regimens, regarding dosing, regarding other questions that we do need to face that will

help us determine what we should expect sponsors to do with them beyond safety assessments.

DR. AUCHINCLOSS: You have suggested that there is a very big difference between this and xenotransplantation. In this case, the vast majority of the immunosuppression protocols will be ones that, in some variation or form, been tried in probably thousands of other patients. Presumably, your safety and efficacy data can be obtained from other organ or tissue types of transplants in humans.

You also have 405 patients who have received islet transplants and you know, I believe, a certain amount about the safety of islet transplantation simply as a technical procedure. It is becoming very unclear to me where any issue data would become terribly useful to the FDA in those circumstances.

MR. SIEGEL: There are a lot of areas in which the safety data for things being tried including the safety of some of the not just immunosuppression but the actual products themselves, more data are needed.

I also do want to qualify that remark about rationale which is to say we recognize the critical importance of these animal models to drug development, to develop the hypotheses to figure out how to optimize a therapy, which ones to take forward into humans.

We are not suggesting that they should not be done

or that they are not useful, simply that, in terms of the amount of that work that will require, or the amount of success in an animal model that will require before human experimentation, that may be limited and will depend largely on the relevance of the model and the risk to the patient and the nature of the patient population because where there are more risks, there needs to be more rationale.

But where there are not relevant models, we don't expect the impossible or irrelevant data.

DR. BLUESTONE: It seems to me that one intersection, though, is a drug that has been tried in another setting in islets as a target potentially for toxicities associated with the drug so that one might think that it might be appropriate to have some kind of safety data, something like human islets transplanted into the SCID mouse being treated with these drugs just to make sure that that drug doesn't have an adverse effect on the islet function, itself, like if you gave steroids to a non-SCID who got human islets, would the steroids be wiping out those islets.

So that may be a model. It is not an efficacy model. It is not even an immunological model. But it is a safety model for how your drugs might interact with the islet transplant.

DR. AUCHINCLOSS: I agree with that. I think that

makes perfectly good sense. You can certainly imagine some very specific questions of safety that the FDA would be interested in what the data is. But they became much smaller questions as I heard their real focus.

DR. CHAMPLIN: I am not sure how reliable animal models are for organ-specific toxicity determination. Often, one sees a totally different spectrum of toxicity in various animal models than in humans although, obviously, any information provides something to look for.

But if something was toxic to the islets in the mouse, I am not sure that necessarily would--

DR. BLUESTONE: No, no; the experiment that I am suggesting is that if we were to say, based on yesterday's experiments, have a mouse model in which you put human islets, portal-vein-inject human islets into a SCID mouse so it doesn't reject them and ask whether your drugs affect the human islets that you put into the mouse--the mouse becomes the vessel, not the islet target.

DR. CHAMPLIN: The only difference, I guess, there would be that you now have a mouse liver that is metabolizing the drug and so it certainly would be an interesting system, again, for screening. But one could envision false results there, too.

DR. MILLER: I just wanted to follow up on Dr. Ricordi's comment that the advances in the Edmonton protocol

would not have been predicted by any animal model. Can you explain to us what animal models were tested to see--because, for those of us who are trying to sort of help to be able to answer the afternoon questions about what animal models do you require, I think if we have had an advance in the field, it would be nice to know why the models did not predict it and whether you think that there are any models that would have helped.

DR. SHAPIRO: We carried out extensive experiments over the previous five to six years in the dog, in autografts and allografts, to try to test what were the optimal immunosuppressive regimes would could apply. We found that the sirolimus was a very effective agent. We knew that tacrolimus at standard dose was fairly toxic.

When we tried our regimen that we now use clinically in that model in the autografts in the dog, it was very, very toxic. We couldn't obtain the data but we predicted it would be useful clinically. We made a big step and tried it clinically, and it worked.

DR. MILLER: How about any of the autoimmune models, the mouse models? Did you look in any other--

DR. SHAPIRO: We didn't. Based on the comments that standard dose or potent immunosuppression therapy is also effective at controlling autoimmunity, we predicted that that drug strategy would also control the autoimmune

recurrence.

DR. SAUSVILLE: Although a point that I would make and, again, we just saw the broad strokes of the regimen. I guess we are talking about a combination of individual immunosuppressives that, in the setting of the human, now has, apparently, been a major leap forward.

It is notorious that combinations of agents, even some of the chemotherapy agents we use, are not well predicted, actually, in any animal model. To that extent, I actually would agree with Dr. Ricordi.

On the other hand, the activity of each of the individual components, at least getting to first base, that this is a reasonable path to begin walking down, would actually revealed in animal models at one level or another at some point in the past.

So I think we have to recognize when the most appropriate time is to utilize the animal information as a determinant of going forward. There, I actually feel the animals are highly valuable in setting these initial safety issues for the first dose in humans.

But I would agree that, for subsequent uses, for combining, there you actually have to build on the more clinical experience.

DR. SALOMON: I think that is a good introduction to Dr. Kenyon who is going to pick up the theme of animal

models and bring us forward into non-human primates.

Non-Human Primate Preclinical Models

DR. KENYON: A lot of the issues I have on the slides you all have already brought up.

[Slide.]

Really, I wanted to point out, too, that if you look in the literature for papers on non-human-primate models of islet-cell transplantation, you will find that there really aren't very many. There are several reasons for this including the fact that, similar to clinical islet isolation and transplantation, it has been difficult to isolate enough islets, viable islets, to get insulin dependence post-transplant.

The drugs that have worked routinely for solid-organ transplantation have not translated well to islet until recently. In addition, as we have already brought up, it is very labor intensive, time consuming and costly.

[Slide.]

I thought Jack did a really nice job of talking about safety parameters and efficacy parameters so I am not going to repeat that. I just need to give you my perspective on the fact that, with regards to the relevance of non-human-primate models to islet-cell transplantation, I think we have to consider that when it comes to safety,

clearly, data generated in monkeys, at least in my opinion, is going to be much more relevant to humans than data generated in mice.

At the same time, we have to keep in mind that the models we are using right now with induced diabetes are generally healthy models, animals. They don't have the underlying physiological changes associated with diabetes.

Then, with regards to efficacy, we have already had intense discussion and I am sure we are going to have some more, that there have been several techniques put forward in mice that prevent rejection, can reverse autoimmunity, and none of those has translated consistently or reproducibly to larger animal models including monkeys and humans.

Another point to keep in mind is really what we are looking at is efficacy for preventing rejection. We are not looking at the autoimmune aspects.

[Slide.]

Then I just pointed this out, that until very recently, protocols that work effectively in mice--I think the mice are very, very valuable. They teach us pathways that we need to address, but in all my experience in dogs and in non-human primates, none of those approaches singlehandedly has worked in a larger animal.

It has been already pointed out that there are

differences in autoimmunity in rodents including, as Dr. Ricordi pointed out, that it is a very explosive onset of autoimmunity. I have been encouraging everyone that I have talked to that works on the NOD mouse to let their mice get diabetes and then put them on insulin and maintain them that way for at least a few months or as long as they can, and then try and do an islet transplantation rather than doing it at the time of onset when you have a superactivated immune system.

[Slide.]

Just briefly, because I do think the non-human primates have a lot of relevance to humans, there are a couple of things, too, that you might not consider that are not necessarily scientific.

The monkeys are more finicky. A dog or a pig will eat just about anything. But monkeys, more like humans, can be finicky about what they eat. It is harder to get them to eat. They are on two feet as opposed to on all fours. In different protocols, not so much for islet allotransplantation, per se, that can have an effect.

Nevertheless, we use primarily a model of pancreatectomy-induced diabetes. It is possible to do a total pancreatectomy in monkeys. It is removed surgically. The down side of that is that it requires enzyme supplementation to replace the lost exocrine function and,

because you don't have completely native exocrine function, it is possible--you can't completely rule out--that the animals have malabsorption and, therefore, don't have the same insulin requirements that a person would with a native pancreas.

Again, this issue that we are addressing, rejection, I think is still a very critical point, though. There is some exciting data recently that tolerance may be possible but, until recently, it really has not been possible to routinely prevent islet rejection.

We can't look at autoimmunity. Again, I want to point out that we are looking at relatively healthy animals.

[Slide.]

With regard to chemical induction, and I know I am expecting Hugh to ask me about this later, it is possible to use streptozotocin. There are a couple of key issues that I would like to bring out. First of all, it is not a non-toxic thing. It is definitely toxic to other organ systems in addition to the beta cells.

When we have done streptozotocin induction, it has been essential to monitor the animals very closely for at least twenty-four hours and up to thirty-six hours, until they clearly stabilize with high blood sugar. But you have to keep them well hydrated to prevent kidney damage.

There is a point where they need bicarb. When the

beta cells start to die, we have to give them glucose for a period of eight hours. So it is not an easy protocol, at least the way that we have done it. I know we will have discussion on that necessarily to follow. We have seen evidence of kidney and liver damage that takes some time to resolve.

Also, with the chemical induction, it is possible to have residual islet function. I think that becomes more important later. I have another slide that I want to discuss that. As Jack mentioned, regeneration is possible, although we did have a monkey that we induced diabetes and kept for over a year and I didn't see any evidence of this at all.

Again, we are addressing issues of rejection as compared to autoimmunity. The animals are relatively healthy in that they don't have the underlying disease changes of diabetes.

[Slide.]

With regards to spontaneous diabetes, and I know we would all love to see a model of type-1 diabetes in monkeys, I want to be very clear here that when I say type 1, I am referring to type-1 autoimmune insulin-dependent diabetes. That would obviously be the most desirable non-human-primate model for transplantation because we could address rejection, autoimmunity and diabetes-related

physiological changes.

However, this is essentially nonexistent in captivity. I spoke with an individual a couple of years ago who has been working with diabetes monkeys for twenty years. He said in his career he had seen one monkey that he was sure had type-1 diabetes.

[Slide.]

So the majority of monkeys with spontaneous diabetes that are reported in the literature are actually insulin-requiring type-2 diabetic monkeys. So they are frequently reported as type 1. They are actually insulin-requiring type 2.

They frequently have significant residual islet-cell function. We actually had four cynomolgus monkeys at the DRI with varying levels of function. You could get different degrees of metabolic control depending on just how much beta-cell loss they had had.

So we are addressing issues of immunological rejection but not autoimmunity. However, if you had a colony of animals with type-2 diabetes, or had access to them, this may be a setting where we can address the issue of safety in the setting of disease-related changes.

[Slide.]

Other key differences in the design of preclinical studies, and I have already alluded to this, is the duration

of diabetes. With our monkeys, we induced diabetes with streptozotocin or we pancreatectomized them, and they get a transplant in a relatively short period of time whereas, and here I am referring to clinical protocols that we have approved at the DRI, our patients that are eligible for transplant must have had diabetes for at least five years

C-peptide and all the clinical trials that I have seen proposed to date, and I obviously have not seen all of them, one of the criteria is that the patients are negative for C-peptide.

If you look in the literature, the monkeys reported are negative in some studies but clearly are present in others. So then, when you are looking at efficacy, you have to try to determine the effect of the islet transplant in a setting where the animal may not have been completely diabetic.

I don't think that necessarily makes it irrelevant, but it is an important issue to consider.

[Slide.]

One of the great advantages of the non-human-primate model is the ability to monitor them very similarly to what we do for humans. We check blood glucose with a glucometer and blood glucose strips, just as we would do for people. You can look at hemoglobin A1c and the first-phase insulin release, in an intravenous

glucose-tolerance test, can be correlated to functional islet mass.

With regards to insulin, a key difference to point out is that humans, fully diabetic humans, need about a unit of insulin per kilo per day whereas the monkeys require 3 to 6 units of insulin per kilo per day.

So, for designing protocols where we are going to look at reduction of exogenous insulin requirement as a measure of graft function, we have to keep that in mind. But one identical finding that we have had, that Dr. Ricordi and Dr. Alejandro and I have discussed a lot, is that when you have a monkey with partial function, it clearly mimics the clinical situation identically.

Depending on the degree of function you have with minimal amounts of insulin, you can maintain relatively normal metabolic control.

[Slide.]

So what are some of the key differences? Jack has already alluded to this. Humans, obviously, when it comes to the organs, the donors and the procurement, in the setting of clinical transplantation, we have variable health status preceding brain death, variable causes of death. The patients have been on life-support for different periods of time.

In the monkeys, they are generally healthy. We do

use, however, older non-human primates as islet donors. They are frequently animals that have been culled from the colony for various problems such as a wasting syndrome in a leg or diarrhea.

But, in general, they are relatively healthy. Obviously, they are sacrificed for the purpose of organ donation and anesthetized at the time of donation. So we take the pancreas out in the OR and walk over to the lab. The only thing that really holds up the isolation starting is me on the telephone.

With regards to surgical technique, the removal is similar. With the variable OPOs, you have different surgeons removing organ. I think someone said yesterday--was it you, Jonathan--that that had been correlated to the islet-isolation outcome.

Obviously, usually, within a center, you have the same surgeon or surgeons removing the pancreas. We don't perfuse the organ because it is not necessary. In the human setting, you have to perfuse it with UW and you have longer cold ischemia times. Ours is generally less than an hour.

So those are key things to keep in mind as we design our trials.

[Slide.]

Then this issue of islet dose keeps coming up. Here, also, we see a lot of similarities. In the human

pancreas, there are about a million islets. I have not been able to find a report in the literature that details that in a non-human primate. If anyone has seen that, I would like to know about it.

It is really clear in the non-human-primate model, just as it is for the clinic, that the number of functional viable islets you transplant is essential and critical to the outcome of your transplant.

If you look at the data that Dr. Hering presented from the International Islet Transplant Registry yesterday, one of the factors that was found to be critical for a successful transplant was a minimum of 6,000 islet equivalents per kilo. Most of the data that you see reported in the literature, insulin independence has been achieved with the use of multiple donors.

Edmonton is now seeing that a minimum of 10,000 islet equivalents per kilo appears to be essential for insulin independence in humans. This is exactly what we have seen in both baboons, cynomolgus and rhesus monkeys at the DRI. I won't anymore do a transplant unless we have at least 10,000 islet equivalents per kilo.

In the literature, people either use multiple donors to achieve enough or, in our case, we will take a larger donor and transplant the islets that we get into a small recipient.

[Slide.]

I was asked to comment, also, on the location of the islets in the liver and the durability. Similarly to the human, islets lodge in the portal spaces with the larger clusters in the portal triads. Insulin-positive islets have been identified in human transplants at five years post transplant. In the monkey, we now have data for insulin-positive islets within the liver at two years post-transplant.

[Slide.]

Also, the issue of matching has come up. It is clear that we really don't know the answer to this question; should we or should we not match? Obviously, matching favors engraftment and prevention of rejection. However, matching may also favor recurrent autoimmunity in the setting of type-1 diabetes.

There are typing methods available for non-human primates. The rhesus is the most well developed. David Watkins in Wisconsin, Judy Thomas in Alabama, have done a lot of typing. Dr. Gaur in Washington State is working on the cynomolgus monkeys; others as well.

So I think that, as time goes on, we will see more and more reagents become available for that. So it is possible to type these animals and look back and see how class I and class II mismatches played a role.

But, again, our clinical trials do not require MHC matching and we have actually made it more difficult for ourselves by purposely choosing the most mismatched animals we can find to look at our tolerance-induction protocols.

[Slide.]

The issue of immunosuppression has come up, as Jack mentioned, similar to the dog, the levels of FK506 and cyclosporine that are needed to prevent rejection of a solid organ in non-human primates is clearly higher than that what is needed in humans.

Now, with rapamycin, I am really not sure. I think Dr. Bluestone and Dr. Hering have a little bit of experience with this drug in the setting of islet transplantation but that is going to be something that we will all be looking at and shows a lot of promise, based on the Edmonton data.

But I think one clear advantage is that many--not all, and Dr. Ricordi specifically mentioned CD3 and the CAMPAC CD52 antibody, but many, many of the humanized monoclonal antibodies that are available for clinical development do cross-react with non-human primates. So if you can do your preclinical studies with these agents, you can get, I think, some nice extrapolation to the clinical setting.

Also, the anti-thymus-site globulins that are used

clinically are clearly cross-reactive in the non-human primate.

[Slide.]

With regard to functional assessment of the islet grafts, I have already touched on the fact that it is possible to do the same types of studies in non-human primates as in the clinic; blood-glucose monitoring, hemoglobin A1c, fasting-plasma glucose insulin and C-peptide, and we can also use identical methods for functional capacity, intravenous glucose tolerance testing, arginine and glucagon stimulation

It is also possible to do clamp studies in non-human primates although there are very few people, I think, that can do that and I am certainly not one of them.

[Slide.]

So, really, I think, the issue that we were discussing in detail before I got up is when is it necessary to generate preclinical data in support of clinical protocols. From my experience, and what we have had at the institute, I would strongly argue that the most relevant time for the non-human-primate studies is when we have novel immunotherapies, antibodies that cross-react, approaches that we can take to show some efficacy for prevention of islet rejection, I think when it comes to using altered doses or combinations of conventional and newer

immunosuppressive drugs, that it is really going to be a case-by-case study because, in some situation, as has already been brought up, the drugs may have been used extensively in other settings and so there is some safety and efficacy data.

[Slide.]

So, obviously, and I feel like I am just summarizing what you have already discussed, the critical needs, if it was possible, would be to develop a model of autoimmune diabetes in non-human primates.

I, personally, also think that we will cure humans before we come up with that. The only work that I am aware of was published in abstract form from Gaur, Nepam and Lernmark in Seattle. They are working on a low-dose streptozotocin model of diabetes in non-human primates.

I think another critical issue is the whole efficacy/safety thing. We have prevention of rejection, but then there is the issue of when you use this drug in an animal with underlying disease, does that make a difference.

It would be possible to look at that, although somewhat impractical, because you could induce diabetes and then maintain a animals for up to a year which is what it takes to see some of the underlying kidney and vascular changes, or we could work on transplanting animals with type 2; for example, treat them with streptozotocin to make them

fully diabetic and then transplant them.

Then something that hasn't really been discussed much and isn't the focus of this session, but, obviously, these models could be very critical for developing markers for testing assays that could predict rejection and help us more favorably protect our islet grafts.

[Slide.]

Basically, what I would like to show you now--we have been talking about all these models and the theories and the concepts. So I just wanted to give you an example of how we have used the model at the DRI. So we have not used tissue typing to date. We are starting to do that in collaboration with other investigators, but we have used a mixed leukocyte culture to identify strongly alloreactive donor recipient pairs.

That consists of taking leukocytes from the recipient and stimulating them in vitro with irradiated donor cells. Then you can look at the proliferative response of the recipient blood cells to the irradiated donor. It gives you an assessment of the degree of alloreactivity.

I am frequently asked, how can you know that this really correlates. In the work that I have done previously, in the dog model, the MLC data clearly correlate with your ability to prevent rejection. If you use low-dose

cyclosporine in a dog, the animals will reject in seven to ten days if they are alloreactive.

If you take MLC non-reaction pairs of dogs, they will keep the graft for 30 days with low-dose cyclosporine as compared to seven in a highly alloreactive pair. So it is efficient for predicting rejection or alloreactivity. In any case, we remove the pancreas from the donor and isolate the islets on day -1.

In our studies so far, the islets have been cultured overnight. And then, in this particular study, we were using anti CD145 from Biogen to test its ability to prevent rejection. So, on day 0, the recipient's pancreas was removed and it was given an intrahepatic islet-cell transplant.

Here is a key difference. In humans, we are using X-ray and a catheter to do the percutaneous trans-hepatic catheterization. In our monkey model, animals are reopened so we put a catheter in the portal vein leading to the liver and drain the islets in.

Jack mentioned the different issues that have to be addressed in islet transplant. We included anti-CD154 in the islet transplant because, in our thinking, it might prevent some of the early nonspecific events that can lead to early islet loss. That clearly has been seen in the monkey model.

Maybe this is a good place to bring up something that Dr. Black mentioned the other day. There is a learning curve. We were talking yesterday about clinical transplant and how many isolations do you have to do before you can do a clinical transplant.

We clearly have that same learning curve in the preclinical studies. Our initial studies were in a baboon and when we switched over the rhesus monkeys, we assumed that it would be essentially the same. But the islets were much more fragile and our first several transplants actually yielded primary nonfunction because we didn't have adequate viable islets.

Once we resolved that, we can consistently now, and routinely get, insulin independence in our monkeys so that the quality of the islets is obviously very important. There is a learning curve.

So, day 0, the recipient is pancreatectomized and given an islet transplant. Then we monitor the monkeys by blood glucose. We look at fasting two to three hour post-prandial and evening glucose.

[Slide.]

We are trying to work very closely together in whatever way we can to design our preclinical studies so that they mirror exactly our clinical studies. So one slide that I don't have, we do the glucose-stimulated insulin

release and the viability on the islets.

We have done over twenty monkeys this way, now. In general, if the stimulation index is over 1--usually, it is higher than that, but I have had one animal that had a stimulation index of 1.2 and the islets functioned very effectively.

The only case I have seen where the assay actually may have really predicted that the islets would not work was in a case where we used two donors. Even though we had gotten enough islets for transplant, it was not insulin independent. When we got the results back of the static incubation, one of the preps was less than 1.0 in the stimulation index.

So I am not sure that the degree of the stimulation index can help us, but it may be possible, retrospectively, to say that it has some relevance. But we do periodic physical exams. The monkeys' weights are taken. Every other week, we have a fasting plasma glucose C-peptide and insulin. This is in addition to the daily monitoring; periodic intravenous glucose-tolerance testing; complete and differential blood-cell count and, because we are using immunomodulators and antibodies, immunophenotypic analysis of the white blood-cell subsets.

In our hands, we have done pre- and post-transplant mixed leukocyte culture to see if the animal

becomes specifically nonreactive to the donor as compared to an unrelated third party.

We look for the development of antibodies to the donor. We do an extensive array. We do P18s for the serum chemistries and, also, in this case, using 5c8 from Biogen, we were looking at 5c8 and anti-5c8.

[Slide.]

So this isn't as relevant to the model but just to explain what we did. We had induction therapy, 20 milligrams per kilogram with Hu5c8 on post-operative days -1, 0, 3, 10 and 18. Then we initiated a maintenance therapy starting on post-operative day 28 and the animals were given a monthly injection of the antibody.

[Slide.]

So just to show you a little bit of what I have been talking about. These are the results that we get when we take a rhesus monkey, do a pancreatectomy and give an islet transplant in the absence of any immunointervention. Fasting blood glucose is the green line. Post-prandial is a purple-pink line. This is milligrams per deciliter on this axis, and this is the post-operative day.

This particular animal, and we have seen this now on several occasions, the post-prandial blood glucose became elevated on post-operative day 6 which, in our experience, has been indicative of rejection.

We initiated an insulin therapy on this day. The fasting glucose started to rise later on, like around day 8 to 10 and what you can see is, even though we initiated insulin therapy with three injections a day, when the animals are fully diabetic, it is exactly like a human. It is very difficult to maintain a normal metabolic control.

This animal was C-peptide negative by post-operative day 10, so the islet were very rapidly rejected.

[Slide.]

In striking contrast, using the anti-CD154, these are the first three long-term monkeys we had. We had long-term graft survival. Antibody was discontinued at about one year post-transplant in these monkeys. All three of them did eventually experience rejection, but you can see there is excellent metabolic control.

Yesterday, Dr. Ricordi showed the slide with a child with type-1 diabetes showing the glucoses all over the place.

[Slide.]

This is just to show the post-prandial glucose. Since anti-CD154 does not suppress islet function, we have actually been able to determine that rejection may be occurring by looking at post-prandial glucose. It elevates before the fasting.

[Slide.]

So how can we use these metabolic assessments, and I promise there are only a few more, to study the monkeys like human? The animals were given intravenous glucose and then the glucose, insulin and C-peptide response was followed after the injection of the glucose which here is at time 0. This axis, as Dr. Hering pointed out to me last night, is incorrect.

But here is time 0 where the glucose is injected. You can see the green line is pre-transplant and then these are the postoperative days. At one year, is the blue line here. What we have seen in our hands is that even in animals with partial function, the glucose response is not an adequate indicator that you have lost functional islet mass.

It can be superimposable even in an animal with partial function but once it has fully rejected the graft, we always see a clear difference.

[Slide.]

What we have found to be much more informative, it has been shown by others that first-phase insulin release in an intravenous glucose-tolerance test is correlated to functional islet mass. We have seen that in our studies.

This is the same monkey that I showed in the previous slide. This is insulin on this axis and time after

injection of glucose at 0 here. The green line is the first-phase insulin release before removal of the pancreas and before the islet transplant.

Then, in the early post-transplant period, the light purple lines, you can see that the first-phase insulin release was blunted as compared to prior to pancreatectomy. Now, the islets do have to revascularize and reenervate and this could be a reflection of that.

However, the animal was insulin independent and normal metabolic control. Then, strikingly, at 155, 227, 296 days, which are represented in yellow here, we actually saw an improvement. One year post transplant is represented here by the blue line. We have seen that in all of the monkeys, that if we can prevent rejection, they actually come up to prepancreatectomy levels at one year post-transplant.

Then the antibody was discontinued at one year. It experienced rejection at 498 days. 539 days, we did an intravenous glucose-tolerance test showing that it had fully rejected the islet graft. And then we did do a retransplant. You can see that we were able to fully restore insulin.

[Slide.]

This is just to be complete, the C-peptide data for the same monkey showing the pretransplant in the green,

the early time post-transplant and the midpoints here in the light purple and yellow and then, at rejection, you can see there is clearly no function left. And then the red line is after the retransplant.

So I think that these models can very effectively give us information on pathways that may lead to prevention of rejection and it can tell us as lot about the survival of islets long-term in the liver if we are able to prevent rejection because that is still a question we frequently get asked.

Thank you.

DR. SALOMON: Thank you, Dr. Kenyon, for a excellent presentation.

I need a sense of the committee. It always sort of comes to the chair to try and stay on time but everybody also wants to make sure that it all gets done. We could have some discussion now of this which, certainly, there is no question this is important area and hold Dr. Eggerman's presentation to right after lunch, which would be my preference. Hugh; you are shaking your head.

Why don't we have some discussion now, invite some discussion, on all these issues including non-human primate and then, if we finish before 12:00, we will just break before 12:00.

DR. SHERWIN: Just a quick question. Do non-human

primates express class II in beta cells? In mice, class II is not expressed in islet cells whereas, in humans, it is thought that it is and, perhaps, that is one of the differences between the two that may have some impact.

Do you know anything about that?

DR. KENYON: We haven't actually done studies with the non-human primate islets. It is a very good point. I have done a lot of studies, some years back, with human islets. I would guess that they are similar. And we didn't see class II in the human islets unless you treated them with cytokines. Then there was clearly dramatic upregulation primarily on the endothelial cells and that is something we should look at.

Hugh, have you looked at that at all? It is a good point.

DR. BLUESTONE: I think the answer is yes, that, at least cynomolgus upregulate class II and endothelial cells in response to gamma interferon, at least. I don't know whether it is the same as human, but it is not exactly like the mouse.

DR. SHERWIN: The endothelial cells within the islet, they are coming from the patient or the recipient. Do you know anything about perfusion? I am just curious now about perfusion of islets. Normally, I assume that they are coming from the recipient. The islet gets revascularized?

DR. KENYON: Correct.

DR. SHERWIN: The perfusion of a normal islet goes straight through the center and then percolates outward so that there is sort of a unique kind of perfusion system for the islet. I wonder if you see the same thing in a grafted islet, would the perfusion go out-in? Has anybody looked at that?

DR. KENYON: That is a good question. I have seen papers on the microcirculation--one paper on the microcirculation in rhesus islets, but it was in the native pancreas. We haven't looked at that.

DR. BLUESTONE: The only potentially relevant point is we tried to do some studies a couple of years ago where we were using adenovirus with beta gal to try to get stuff into islet. We injected it into--actually, this was when they undid the kidney capsule and actually found most of the blue cells were around the outside.

DR. SHERWIN: That is what I bet. So there may be some things about islet physiology that change during transplantation that could influence results. I mean, I have no clue as to how.

DR. SALOMON: It is interesting to go back to where we were yesterday where we had a fairly low opinion of the necessity to look at glucagon- and somatostatin-producing cells yet today we are sort of taking

the opposite tack that this may be important in terms of regulation of the--I am just pointing out the--

DR. RICARDI: Well, it may be important in terms of research but I think one of the important findings with these metabolic studies is that traditionally it was thought that an islet transplant could not reproduce a pattern of first-phase and second-phase insulin release and has this typical blunted first phase with delayed response in insulin production.

What the study shows it that clearly in the large-animal preclinical model that actually this is an issue related mainly to islet mass transplant that survives because if you have sufficient islets, you have a pattern that mimics exactly what you would expect in a more physiologic condition and actually demonstrates, also, the ability of islets to improve in function over time in the absence of any rejection or autoimmune recurrence problem.

DR. BLUESTONE: Norma, I have a question. One of the things that the large animal models offer that the smaller ones don't is to look at where the islets go and migrate. You talked about the liver. At least it has been our experience that we find islets all over the place. We find islets in the lung and stuff, and we probably don't inject them as well as you do.

But I am just wondering, because the notion that

the liver would be a good site for this because of immune privilege and things like that--have you looked? Is it possible that some of the long-term variability and success depends on where the islets go as much as how good the islets are?

DR. KENYON: When you say "long-term variability," what are you referring to?

DR. BLUESTONE: I am talking about long-term insulin independence of human beings after giving the islets.

DR. KENYON: That is a good point, Jeff. We have not had a lot of tissues that we have started studying extensively, but we have not looked for them in other places. And I will. It is always a possibility.

Camillo, have you done any studies in the dog looking at other tissues?

DR. RICARDI: There have been studies like injection islet in the lungs as a site of transplantation. But I wouldn't--

DR. BLUESTONE: It was really very striking. We tried to do some biopsies in the liver, as you do. As you know, it is depending on where you biopsy, you see them or you don't. We did two biopsies, couldn't find the islets. We said, "What is going on here," because this monkey does not need insulin.

We were worried. We did the pancreatectomy. Still didn't need insulin. So we just took other organs, including the lung, and we found a lot of islets in the lung.

DR. RICARDI: And they were infusing the portal vein.

DR. BLUESTONE: You bet. Although, I can't vouch for that. I am not the surgeon.

DR. AUCHINCLOSS: Let's start by putting this strep issue to rest. We work primarily with the cynos and I know that is not your primary species, but I know you have also. We find strep treatment to be very consistent, very effective, no regeneration of islets in at least two dozen examples that I can think of.

The cynos tolerate the strep treatment if you hydrate them first. There is some renal toxicity. We get the C-peptides well below one. You clearly see big changes after a successful islet transplant. Blood sugars are clearly abnormal and require insulin and then can be normalized without insulin.

I think that the strep model is an equally acceptable model for islet transplantation in non-human primates; do you agree with that?

DR. SALOMON: Can I also add our experience with rhesus. I would also acknowledge some help initially from

Phil Padrid at the University of Chicago who is a good consultant. We have done it now in about sixteen rhesus macaque monkeys and had essentially the same results.

DR. KENYON: My question, then, would be what are the normal fasting C-peptide levels in your monkeys and how do they compare the monkeys after the streptozotocin because, in my experience now in baboons, rhesus and cyno, not with strep, we have used strep in rhesus and baboon, the fasting C-peptide can be anywhere from 0.8 to 4.0 depending on the monkey.

Even in the published literature, they will show the normal range of the C-peptide and then the normal range of the C-peptide in the monkeys that are getting a transplant, and some of them are in the normal range. So do you any IVGTT to prove that there is no C-peptide release in response to a stimulus.

If I could see that, then I would be totally satisfied and then it is just a matter of us, obviously, working out the logistics because it has been very labor-intensive with the approach that we have been using.

But my main concern is does it really eliminate C-peptide?

DR. BLUESTONE: We routinely do IVGTT before we do the transplants. Our monkeys are usually two weeks out post-strep treatment. I would say, in 80 percent of the

monkeys, we see zero--within the limits of the ELISA detection C-peptide. There are a subset, about 20 percent, that we do see some, anywhere from 0.2 to 0.4, 0.6.

But in the majority of animals, we can wipe out totally--

DR. KENYON: I think if you show that, then it is actually a preferable model because you have the intact exocrine function because they don't like the Viokase very much.

DR. AUCHINCLOSS: The second point I wanted to bring out with respect to your presentation was the HLA-matching issue where I think we ought to be clear that we do not expect to accomplish HLA matching for islet transplantation in the future. That would essentially turn it into the problem of trying to find a bone-marrow transplant for a 6-antigen-matched bone-marrow transplant from a nonrelated individual, in which case, we might has well forget islet transplantation as therapy.

So, to me, the only issue for HLA matching is to make sure that your monkey model is not matched which is, of course, what you were doing with your MLC cultures ahead of time, and the rest of the HLA matching, I would forget about entirely as far as islet transplantation is concerned.

DR. KENYON: I actually agree with you and I should have explained it more clearly. I didn't mean to

match in the setting, if you would, in a bone-marrow transplant. I come from the solid-organ perspective where one DR, or something like that--but even that, we are not trying to achieve. So that was my point, not to match completely.

DR. SHERWIN: I have two questions. I am just curious about matching, just for my own education. You are talking about class I and class II? Does it matter?

DR. AUCHINCLOSS: You won't even know. You won't even look.

DR. SALOMON: The only problem with an MLC is that it is more class II.

DR. AUCHINCLOSS: It is class II, but you can mismatch your monkeys for class I and II.

DR. HERING: Let me ask you, 25 percent of the donor population is haplo-identical with type-1 diabetic patients so it would not be completely inconceivable to find a haplo-identical donor for a type-1 diabetic recipients. Would you think that could have an impact?

DR. RICARDI: Would that increase the possibility of autoimmune recurrence?

DR. AUCHINCLOSS: I suspect the answer to Camillo's question is yes. I think the answer to your question is probably yes, it would have an impact. My suspicion would be that the impact would be so small as to

be absolutely insignificant compared to all else that we need to do.

DR. BLUESTONE: I don't think the first part is true. So if I had to predict what happens is that endothelial cells, which come from both--probably something from the donor, but also the recipient which are localized there--will reprocess peptides and present them in the context, whether there is a matching or a no matching, and trigger release of cytokines locally which cause damage.

I don't know any reason to think that direct recognition by class II cells is going to be the major pathway to destruction here.

DR. SHERWIN: I would totally agree. The question I was really getting at was class I, which is a different story. I just think it is important to think about. I think class II matching is probably not as important as class I--and to look at those issues; I think that is important.

My other question really related to the liver, itself. Have you looked at what the liver looks like metabolically? When you put an islet into the portal vein, the levels of insulin around that islet are going to be astronomical, as they are within the islet, many, many logs higher.

So, presumably, around the islet, there is a lot

of glycogen. That question is how have you changed the liver in any way in the area of the islet? Have you looked at that?

DR. KENYON: We have done some initial assessments. When we sacrifice monkeys with partial function, the intact islets actually don't appear to have any deposition around them. But we haven't done the staining yet. We have really just started analyzing all the tissues. You see some lymphocytes if the animal is undergoing rejection.

DR. SHERWIN: I guess the only issue to think about down the road, and may probably not be an issue, is those kinds of extremely high concentrations could be growth factors. So, it could theoretically lead to tumors or things like that. It is surely something to consider, even though I am not saying that there is any evidence to support that view.

DR. KENYON: Sure. But, also, with regards to the liver function, we do look at liver-function tests every other week. In the immediate post-transplant period, you will see an elevation in some of the enzymes, but then they resolve within a week.

DR. AUCHINCLOSS: I have another more general question for the committee and for the FDA. A lot of people are talking about the animal studies but particularly, with

the non-human primates, we are talking about the efficacy of various drug combinations or antibodies, et cetera.

My question is, to what extent do you feel that these tests have to be organ- or tissue-specific. I would have suggested that 98 percent of what you learn from a kidney or a heart transplant in a monkey with CD154 or reagent of choice is transferrable to islets, as well--not 100 percent, but 98 percent. Do you agree?

DR. KENYON: No. It hasn't been our experience. Traditionally, things that have worked for solid-organ allografting in primates, including conventional immunosuppressive drugs, have not worked for islet. Especially, in our hands, FK hasn't. We haven't tried rapa yet. But, no; I don't think it is 98 percent.

Some of the newer things, the CD3 immunotoxin being a prime example, is an exception.

DR. CHAMPLIN: If you think about vascularized grafts and cellular grafts with kidneys on one end and a bone-marrow transplant on the other, certainly what works in kidneys doesn't work for bone marrow and there is very different immunosuppressive drug requirements for that type of transplant.

So we were chatting whether an islet is a tissue or a cell transplant. It is a small tissue, I guess--lots of them. So it may very well have some unique

characteristics and I wouldn't, necessarily, assume that things would cross over from solid organs.

DR. CARA: I am sorry for this very sort of basic question, but I need some education in terms of some of the phenomena that you are talking about. How is the immunopathology, if you will, of rejection different from the immunopathology of type-1 diabetes and is it important to know the difference if you are going to be using therapies designed to suppress rejection, perhaps even autoimmunity, during islet-cell transplantation?

DR. BLUESTONE: Good question. There has been very little--if you want to get back to animal models like the NOD mouse, we could talk a little bit. But, certainly, in human beings, we have very little information about what--there are questions, still. People argue that antibodies are not important. I think there is no data in human being as to whether the antibodies are important or not.

The issue about relative role of class-I versus class-II-specific cells, and stuff, I think it is an open question. There is enough controversy in the mouse model. The human disease, I think, is really a totally open question.

DR. SALOMON: I think that is what was part of the conflict earlier about even how relevant those two

mechanisms are.

DR. RICARDI: In part, you may consider that in an allo-reaction, you would expect destruction of the entire islets with an autoimmune kind of immune attack. You would have a selective beta-cell damage. But this is actually more complex than that because if you have a failing islet autograft, you can find selective persistent alpha cells alone and selective loss of beta cells.

So, because of the sensitivity of beta cells to cytokine damage and other problems, you can find something that mimics an autoimmune kind of islet destruction even in autotransplantation or in allotransplantation. This is a very difficult issue to be addressed.

Regarding the change in the liver, there are, indeed, some early changes, even if you follow liver-function tests, there are normalized very soon after islet infusion--there are some early changes that you can find in animal models that is just the peri-islet row of hepatocytes in which you can see glycogen deposition.

As a matter of fact, the way to find islets at low magnification, you just look for a glycogen around the liver and then you zoom in to get the islets in the rodent models. But this seems to disappear with time and revascularization and we have limited experience.

But, in the clinical setting, like long-term, the

five years or nine years islet functioning in human livers and in late biopsies, you will see pretty intact, what we can say, hepatocytes around the islets and no late sign on liver function.

But I agree that it is a field that could be investigated more carefully.

MR. SIEGEL: You mentioned in the baboon, cyno and rhesus, not much dissimilar to humans, you needed about 10,000 islet equivalents per kilogram. Do you have data regarding whether, for a given cell number, the viability of the prep or in vitro functionality or the size distribution of the particles in the animals are predictive of success?

DR. KENYON: Yes; Dr. Black asked me this frequently. The 10,000 number, basically, we came up with based on experience. That is the number of islet equivalents that we can give and consistently and reproducibly get insulin independence for the first week regardless of what they are treated with.

With regards to where they go and is that predictive, those are things that we are trying to address now with a lot of the tissues that we have. I don't have the answer yet. I think the functionality part, we have looked at in vitro glucose-stimulated insulin release in twenty preps that got transplanted.

Unfortunately, when we had our initial learning

curve, we were not doing those tests because I might be able to answer your question. But now, the majority of the transplants work. The one that did not work, the animal had islets from two donors. So I was surprised, because it clearly got enough islets.

But when we got the results of the in vitro studies, one of the preps had no stimulation at all and so probably wasn't good. So it does appear from the very limited experience that we have that we might be able to make a little bit of a correlation.

But the actual number of the stimulation index, if there is stimulation, I see a range of stimulation indices in successfully transplanted animals. So the only correlation I can draw right now is that, in a prep where it was actually the stimulation index, that animal didn't become insulin dependent.

But I don't think it is a high enough N to have an impact on the clinical--

DR. RICARDI: These were also done after culture for one day overnight, so they are not fresh, they are not immediately transplanted like in the Edmonton protocol.

MR. SIEGEL: Just one quick question on point of fact; you mentioned that the intravenous glucose-tolerance test, the first-phase insulin release, if I understood, was correlated well with the functional islet cell mass.

You said that earlier in your talk. Later, you showed how that variable seemed to vary over time. Were you referring to the results that you obtained immediately post operative, or the results a week later or a month or year later, or what?

DR. KENYON: That point has actually been shown by other investigators, primarily Paul Robertson, that there is a correlation between the functional islet mass and first-phase insulin release.

But, interestingly, what we see in the first couple of months post-transplant, the height of that first phase is correlated to the number that you transplanted whereas, later on, over time, as the islets revascularize and settle in, at one year post-transplant, they seem to come together and achieve their pre-pancreatectomy levels.

So, other than the fact that I see a correlation in the immediate post-transplant period--for example, one monkey got 40,000 islet equivalents per kilo and it actually had an insulin release post-transplant that was much higher than pre-pancreatectomy. So we, appropriately, named that monkey Camillo.

Our monkeys that get less than--the one that I showed you that had the blunted first phase at 42 days only got about 11,000 islet equivalents per kilo. So you can see the first phase marching up. But then, over time, they come

together and it is not as indicative.

DR. SALOMON: I think, at this point, we are five minutes after the afternoon and I would like to stop. I know some of us have to check out. This is an excellent discussion from some excellent presentations this morning. I want to thank all the speakers.

These are the issues we will be discussing the rest of the afternoon, so I don't see any big issue to stop here. So I would like to have everyone back, if you don't mind, at no later than 12:45 so we can get started on the afternoon meeting.

[Whereupon, at 12:05 p.m., the proceedings were recessed to be resumed at 12:45 p.m.]

AFTERNOON SESSION

[1:05 p.m.]

DR. SALOMON: I would like to get started with the meeting with Tom Eggerman from the CBER staff who is going to sort of give us an introduction into some of the questions we want to deal with this afternoon.

I did want to tell everyone that we have had some discussions just practically looking at when most of the members are leaving to go to the airport, including me as the chair. I was going to delegate it, but everyone I delegated to is also leaving at 4 o'clock.

So, after discussion with the FDA, what we are going to do is actually stick to finishing this meeting a few minutes before 4 o'clock. I hope that doesn't require me to cut any important discussion off, but I think if we can try and make clear, sharp comments and get all the discussion in, I think that will be better for everyone.

Okay, Tom. You're on.

FDA Perspective, Clinical Issues

DR. EGGERMAN: Good afternoon.

[Slide.]

My name is Tom Eggerman. I am with the CBER Division of Clinical Trials Design and Analysis as well as the Division of Cell and Gene Therapy. I would like to discuss with you issues in early clinical-trial development

of allogeneic islet therapy from the FDA perspective.

[Slide.]

As was excellently presented yesterday, islet therapy has been developing for over fifteen years. Over this time, there have been a limited number of patients who have been treated in a number of centers throughout the world and also, over this time, the technology for producing islets has been refined, as have the clinical immunosuppressive approaches.

Many potential sources for islets have been evaluated including both fetal and non-fetal allogeneic, autologous and multiple xenogeneic species. A few islet therapies have been associated with devices, both encapsulated as well as macro-device-associated technologies to help address the problem of immunologic rejection.

In today's discussion, we are focussing on allogeneic non-fetal pancreas sources for islet therapy. We will be concentrating on the issues associated with early clinical-trial development, especially addressing the safety and activity assessments. Even though this therapy has been used for over fifteen years, the limited successes have not allowed trials to really advance beyond phase I safety studies.

Yesterday, some very encouraging data was alluded to that will, hopefully, eventually translate into pivotal

trials that will truly evaluate efficacy as well.

[Slide.]

An important aspect of the evaluation of the safety of proposed clinical studies submitted to the FDA is evaluating the eligibility criteria to determine if there is an acceptable risk/benefit for the patient population that is being studied.

In protocols submitted to the agency, the eligibility criteria have included patients with type-1 diabetes, with advanced disease. The specific criteria have included a negligible endogenous C-peptide level, a history of diabetes for at least five to ten years, a history of poor glycemic control including a number of documented hypoglycemic episodes and an elevated hemoglobin A1c.

Most trials have enrolled patients who are already under immunosuppression related to previous organ transplantation, usually kidney. Some studies have specified tissue matching such as ABO or HLA.

[Slide.]

In most studies of allogeneic islets submitted to the FDA, patients are on concomitant immunosuppression which is associated with well-known risks including infection, nephrotoxicity and neoplasm. For those patients already on immunosuppression for other organ transplantation, there is not the added risk of new immunosuppression but it is

recognized that this may not be the optimal therapy for islet transplantation.

Some investigative studies are enrolling patients for islet therapies that are using immunosuppressive regimens specifically designed for optimized islet therapy. Yesterday's presentation by Dr. Shapiro illustrated this approach.

In view of the risks associated with immunosuppressive therapies, other techniques have been developed to minimize or eliminate the need for immunosuppression. These have included devices to immunoisolate the islets, the development of tolerance procedures and the use of epitope masking procedures.

[Slide.]

FDA evaluates general safety for the entire therapy including the procedure used, the islet product as well as any concomitant therapies such as immunosuppression. Routine evaluations include clinical lab monitoring such as CBCs, chemistries and urinalysis as well as follow-up clinic visits.

When there is concomitant immunosuppressive therapies, clinical and laboratory assessments are preformed appropriate for the specific regimen. In addition, the clinical protocol includes predetermined stopping rules which require the cessation of patient enrollment for the

development of severe or clinically significant toxicity.

[Slide.]

Because of the specific disease aspects of diabetes, the safety assessments also include diabetes-specific monitoring which include patient glucose diaries, the number of hypoglycemic episodes, hemoglobin A1c and other gylcated proteins.

Some trials have also monitoring anti-islet and/or anti-insulin antibody titers and, since islets produce other proteins, there has been consideration of evaluating antibodies to these other proteins as well.

[Slide.]

Unlike many products, defining a dose in islet therapy is not so straightforward as was discussed yesterday. There is a disagreement as to how a dose should be defined. The two methods most commonly used are related to the number of volume, so-called islet equivalence, or reflect an in vitro islet function prior to administration.

Hopefully, with more standardization in the field, the best method will become clear and may reflect some combination of these two elements. In most studies, a single administration of islets has been evaluated. Some studies have used sequential administrations to reach a predetermined dose.

Over time, islet function and/or number will

likely diminish requiring a second administration of islets to maintain a certain level of islet-related insulin production.

The optimal timing for second and subsequent administration and the potential success for second and subsequent administration remains to be determined. If allogeneic islet therapy becomes successful, source limitations reflecting the limited number of potential organs will greatly limit the use of second administrations since the number of potential diabetes patients greatly outnumbers the number of organs donated.

It is hoped that advances in cell culturing, genetic engineering and stem-cell biology will eventually allow either the expansion of islets of the establishment of expandable pools that would allow the production of unlimited numbers of islets so that all patients could be treated initially when appropriate and then retreated when necessary.

Alternatively, sources such as xenogeneic islets offer a relatively unlimited supply but raise other potential infectious disease and immunologic issues.

[Slide.]

A concern has been that immunosuppressive regimens that have been developed for transplantation of organs such as kidney may not be optimal for islets. As was presented

yesterday by Dr. Shapiro, one approach to optimize islet survival has been to develop islet-specific immunosuppression.

Many believe that elevated glucose levels, particularly at the time of islet therapy, can be toxic to islets and an approach for this potential problem has been to use tight glucose control immediately before, during and for a period after islet therapy.

[Slide.]

The most commonly used route of administration has been injection into the portal vein. However, there have been serious adverse events associated with this approach which is intended to reproduce the normal insulin secretion which is transported through the portal vein from the pancreas.

Other sites have been used which are usually associated with a device, primarily subcutaneous and peritoneal sites. The advantages of these sites include the ease and decreased risk of administration and the ease of product removal to better understand the survival of the islets or to remove the product if there was an adverse event associated with its use.

[Slide.]

The informed-consent documents include a discussion about potential islet-therapy procedure risks,

potential infectious-disease risks as well as the risks of any concomitant therapy.

The consent process also informs prospective study participants of alternative therapies, of the potential risks of alloimmunization including the potential negative impact upon subsequent organ transplantation as well as repeat islet therapy.

[Slide.]

There have been multiple potential outcome measures of activities that can be determined for islet therapy. These include glucose diaries, measures of glucose variability, fasting glucose levels, hemoglobin A1c or other gylcated proteins, insulin usage and C-peptide measurements, either basal or stimulated. One of our questions to this committee is whether levels of other islet proteins should also be determined.

[Slide.]

When islet therapy advances to the point of pivotal trials, a major question will involve appropriate efficacy endpoints. If possible, insulin dependence would be desirable. However, other outcomes such as improved glucose control, may be a potential efficacy endpoint. This may be particularly important in brittle diabetics with hypoglycemia unawareness and a history of life-threatening hypoglycemic episodes.

Other clinical endpoints such as retinopathy, nephropathy or neuropathy that reflect improved glucose control may also have the potential to demonstrate meaningful benefit. A question that will also need to be answered is what durability of efficacy would be clinically meaningful.

These are examples of many important issues and islet therapy. We look forward to your insights and perspectives this afternoon.

Thank you.

DR. SALOMON: Thank you very much, Tom.

Committee Discussion--Preclinical/Clinical Issues

DR. SALOMON: In preparing for the meeting, I had several discussions with FDA staff. I wanted to start off, then, this last three hours or so of the meeting by trying to do justice, very briefly, to what the FDA staff wanted to get out of this meeting.

Yesterday, we identified a series of issues that relate to identifying and assuring the quality of the product which is extremely important in terms of thinking for a regulatory agency. Again, I think the message the FDA is trying to get to the field is that, by doing this proactively instead of reactively, would be to emphasize to everyone that they want to be a partner in the development of this moving forward and not create product criteria in a

vacuum that would, in any way, impeded progress.

From product to preclinical to clinical is kind of where we are going now. For the FDA, they were very insistent that I get the message and stay on track in the discussion, not to jump to clinical so far that we don't deal with the implications of preclinical models, to the extent that we believe in preclinical models, because we have already had some discussion of those issues and they need to be on the table this afternoon.

So when we are talking about clinical-trial designs, the FDA wanted to always come back to what kinds of questions can be validly answered in what animal models because, once again, when the question comes up to initiating a particular kind of IND-based proposal, how much safety has to be demonstrated preclinically in an animal model so that the FDA feels reassured and that the public, obviously, is reassured that we have done our diligence.

So I think these are sort of the key questions. If we can kind of keep that in mind and remind ourselves to comment on the preclinical models at each juncture, I think we will be serving the FDA well.

So I would like to begin this series of questions, at least initially, in order. The first question is that of immunosuppression.

DR. AUCHINCLOSS: Back to the issue of why is this

conversation that is about to take place going to take place at all. I have been rereading your proposed approach to regulation of cellular and tissue-based products from 1997. If I understand it correctly, you feel that regulation of islet transplantation is appropriate because there is a metabolic component to the tissue but that if this were an autologous islet transplantation, regulation would not be required except for the process of islet preparation.

Is that correct?

MR. SIEGEL: If it were autologous and not more than minimally manipulated, regulation would not be required, period. Well, that is wrong--as a product and, therefore, much of the process of manipulation, many aspects of it, would not be regulated.

However, your statement is more correct than I initially indicated because it would still potentially be regulated under our authorities regarding transmission of communicable disease which we use to regulate tissues largely vis-a-vis issues of donor testing and screening but also issues of insuring that the processing does not damage the quality of the tissue.

DR. AUCHINCLOSS: So you are interested in the safety of the tissue from infectious-disease point of the organ donor and you are interested in the process of the preparation of the islets, but you would not regulate the

trial use of autologous islets.

And you feel that islet preparation, at least according to this document, does not involve more than minimum manipulation, at least as I read the document, which says, "extraction or separation of the cells from structural tissue," blah, blah, blah, "is not more than minimal manipulation."

So the only way in which I find that you are interested in regulating allogeneic islet transplantation is subsequently you say, "Well, metabolic function; if the cell product has metabolic function, then we want to make sure that it has metabolic function and, at that point, we feel an IND is necessary."

But then you go and you say, "Well, it is not necessary for an autologous islet transplantation because that is going back into the same recipient." I don't understand the rationale for that.

MR. SIEGEL: There is a rationale. It was based on months of back and forth to various regulatory committees and discussion with various groups. But, basically, there is an attempt to draw a number of lines here between what should be regulated as a tissue and what should be regulated as a biological product.

It was generally felt that, for example, tendons or bone chips which are in, at least some sense, not alive

have, in many cases, there is a higher a priori presumption of efficacy.

If you take a tendon and you use it to replace a tendon in an individual, the regulatory concerns about clinical efficacy and appropriate function are relatively small. However, there are a number of factors--and, therefore, the regulatory focus of that guideline for that class of products is on safety issues regarding making sure that it is free of contamination and that it is stored in a manner that wouldn't allow its intrinsic function to deteriorate.

Conversely, you have mentioned two of the three or four types of issues that would cause such a product to require marketing approval. One is when a product is more than minimally manipulated. That involves what we would generally consider manufacture.

Examples might be expansion of cells, genetic transduction of cells. When one does that to a product, while it is hard to draw hard-and-fast lines, it is necessary to draw hard-and-fast lines in terms of telling the world how you are going to regulate things.

You can't just say, "I will know it when I see it." One of the lines that I think is a reasonable line is that it is much more reasonable to presume that something is functional when it is being used to do what it did and it is

being used in its original form as opposed to when it is more than minimally manipulated.

Another factor that you mentioned is a combination of allogeneic and metabolic or systemic activity in allogeneic source. The reason those are combined together is because there is a reasonable presumption--for example, if you take a blood vessel, a saphenous vein, say, or, say, from a patient or you take an ovary out while you irradiate their pelvis and restore that ovary, or you take a vein out and put it into another area, there are reasonable presumptions about clinical efficacy and safety that are not there when one uses allogeneic tissue, both because of issues of rejection but also other issues of biological compatibility.

Again, those are not lines that are certain. You can give counterexamples on either side of those lines where the concerns are higher in one group than in another group but part of the goal here, and a critical part of the goal, is to set out the rules in advance so somebody, when they are deciding how to do their research, how to invest their funds, what to develop, will understand what is the regulatory framework. Without that, significant damage can be done to product development.

A third area is homologous usage. So if you take that tendon and you use it as a tendon, that is one thing.

If you take that tendon and you use it as a ligature to prevent embolization in a vein, that is a different usage. Of if you take that tendon and you make claims for it for other uses such that we can implant this in your abdomen and it will cure cancer or AIDS or something like that, we consider that regulated.

So that is the rationale why either more than minimal manipulation, allogeneic and metabolic function, or non-homologous use are factors that cause products to be regulated as products rather than as tissues.

DR. SALOMON: Jay, I would also question if the fact is, to the extent that anyone around the table accepts the premise that there are immunobiological features that are unique to islet transplantation--albeit there may be some disagreement on the details, I think all of us accept that overall premise--then the use of different drug combinations--in some clinical trials it is going to be new drugs that haven't been tested before.

Certainly, the islet community is very excited about the use of really new drugs that haven't been fully tested. And, certainly, biologics. There is another rationale here, even with established drugs, that there should be some oversight on the design and conduct of the clinical trials to assure the fact that data obtained in the experience of older or newer drugs in kidney or liver

transplantation, let's say, is relevant to what is going to happen in our patients in islet transplantation.

MR. SIEGEL: I think that is moot in the sense--I think that is right in the sense that I indicated. In a large majority of these cellular applications, it would be under FDA purview. We have also talked about encapsulation devices. We have talked about concomitant experimental immunosuppressive therapies.

Although, not at this meeting, we have every anticipation that many of the technologies talked about here will give use and be applied to cellular expansion and genetically modified cell technologies as well as in vivo growth of cells with various regulated factors and products.

So, in most cases, that is the case. But in some cases, it does make a substantial difference whether the cellular product, itself, is considered a regulated product.

DR. AUCHINCLOSS: Please, I do not suggest that I don't want good trials of islet transplantation, that I don't want oversight of those trials. I just don't think the FDA should be the source of that oversight.

Your rationale for regulating islets, when you read the whole thing--you mentioned a variety of criteria. It comes down to the fact that it is allogeneic and metabolic. You don't regulate all allogeneic, do you, because organs evoke an immune response. So it is not just

allogeneic.

It would make sense to me if you said, "I want to prove, when you do an islet trial, that you produced an islet that knows how to make insulin." That would make sense to me. But then it would apply to an autologous transplant just as much as an allogeneic transplant.

So the FDA would be in a great position to help us insure that the islets that are produced in facilities are really islets and that they make insulin. But there your job can stop. Once we show that the islet makes insulin, we can design the clinical trials.

MR. SIEGEL: I should just say--we could debate this forever--

DR. SALOMON: Let's not.

MR. SIEGEL: And I am not sure it is particularly useful. I should say that there are different laws that apply to the area of solid vascularized organs. So your question, they are metabolic, yes. But there are different laws specifically that apply to how those are used in this country.

DR. AUCHINCLOSS: Yesterday, you suggested that if once we called it a product, it had to go the whole distance. But your document here does suggest that there are kinds of products for which you want to insure safety of the tissue and adequacy of the process but that do not

require INDs and premarketing licensure.

MR. SIEGEL: The whole purpose of this structure is to apply the level of regulation as appropriate to the types and nature and extent of issues concerning safety and efficacy raised by the product.

Those products that we do call tissues are regulated predominantly for infectious-disease risk although we believe we have some authority based on the infectious-disease risks to also make regulations pertinent to product quality.

However, beyond that, we don't have options to regulate a product in the ways we are talking about, to just regulate how it is manufactured and stop there and say it has to be able to make insulin, but it is not a product. That just doesn't fit into our regulatory--

DR. AUCHINCLOSS: I don't understand this document, then.

DR. SALOMON: I didn't want to interrupt until now because I think Hugh's points should be a part of the record. If he has concerns about a discussion that now follows, then I respect that from him. I think we have got that in the record, now.

I don't think that the purpose here is for us to debate what decisions the FDA has made on whether to regulate or not, although I think Hugh's points, perhaps,

should be considered in detail by the FDA. So I would like to get back to the topics at hand.

Hugh, are you okay with that? I don't want to deprive you of a key point here.

So, the first thing would be immunosuppression. I think maybe sort of the overriding question on the immunosuppression is what do you guys think would be the optimal immunosuppression to use in an islet transplant. I think we have got some data on the table on that already, but let's put that as a specific question.

What data from current preclinical models justify, in your opinion, that decision. We also touched on that a little bit earlier but let's just make sure that we come to some sort of conclusion on that.

DR. RICORDI: Actually, I would like to make a comment in support with Dr. Auchincloss' previous comment and that is that I don't think it is necessarily our business to discuss the best immunosuppression for islets in this site, but I completely agree on the fact that we should address safety and product-release criteria and what is the best islet that we can put in patients.

But we are here to develop a procedure over the years that is extremely more safe than a pancreas organ transplant, like maybe ten-fold safer as to morbidity or mortality, but in which, as things are going or developing,

would be imposed on regulatory aspects that are ten-fold more complex than what you have to do in organ transplantation.

So I think the message is to go back to full pancreas transplantation from the kind of discussion here. I have to agree completely with Dr. Auchincloss that I am completely supportive of the idea of standardizing product-release criteria and safety concerns but it would be severely damaging to the whole field of allotransplantation, for example, to impose a unified protocol, that everybody now does the same thing for the next two years in a stage where nobody knows which one is the best product development of kind of tissue separation, et cetera.

DR. MILLER: I actually don't think that is really what they are asking. I think they want some framework around which experts feel are acceptable protocols for them to review, how much more information do the experts feel that you need to collect before you can proceed with clinical trials.

You may say that all the information is already there, either from the clinical experience or the animal experience, and that is the question that they are wanting to ask, is what I think we are here for.

DR. AUCHINCLOSS: Carole, that is not true. The question says, "What is/are the most appropriate

immunosuppression regimen(s) to use for islet-only studies?" That is an absurd question.

DR. CHAMPLIN: I guess my fundamental issue or question is what is the role of the FDA in defining these sorts of things as opposed to individual institutions and their IRB. I would agree with the idea of trying to define the product and the safety issues and the product description of things that really relate to the transplant infusion.

But the issues of how best to treat patients, what would be the eligibility criteria for people going on to clinical trials--certainly the big area of immunosuppression is something where one should not try to impose a preconceived standard when there really is so little data of efficacy.

MR. SIEGEL: I think there is a lot of putting up of straw men here to shoot down. There is no discussion of imposing standards. We have a requirement to insure that these trials do not expose patients to unnecessary and unreasonable risk.

In order to achieve that, we need, and we are looking to you, for advice, better understanding how to assess those risks of when and whether additional animal models are appropriate before going into humans, as to which risks are more significant in which populations as a

function of population, as a function of dosing, and, as products develop, we additionally need--in addition to insuring that there are not unnecessary and significant risks, we need to insure that there are data of an adequate nature and quality to be able to assess the safety and efficacy of the product.

Those are what our mandate is to do. We do that in many areas. We don't, in those areas, tell people how to do their clinical trials. We don't tell people that there is only one way to do a clinical trial. We try to reflect the best science and to add our expertise in clinical-trial design, our expertise in what sorts of methodologies work, what sorts of inferences can be made from what types of study designs, add that to the expertise that we receive to help people do trials that will be safe and will be meaningful.

DR. SALOMON: So the question comes if you have a clinical trial that you want to do--let's back up a little bit. I don't want to tread in such sensitive ground immediately. But if there is a clinical trial that you want to do, I assume you want to use immunosuppression or will want to use some form of immunosuppression.

How would you suggest that immunosuppression, at the time you are going to initiate that trial, be justified? What kind of data from a preclinical model would be

reasonable to present?

Jeff, do you have a comment you want to make on that line?

DR. BLUESTONE: No; obviously not, because I was going to ask you what you said. I was actually going try to find a middle ground, here.

DR. SALOMON: I am not saying my middle ground is any better than your middle ground, by the way.

DR. BLUESTONE: So my middle ground would be to ask your question somewhat differently which would be to ask the question, is there anything that we can agree on that is a necessary component of the regimen that everyone should be doing.

I think the answer we are going to say is not and, therefore, it would be in Hugh's category. For something else, the answer may be absolutely yes, like 10,000 islet units which is maybe what we decided yesterday.

I actually don't see the distinction between yesterday's discussion and today, only that we knew more yesterday about what we liked and today we haven't had the discussion yet. I think your question is fine, but my answer to the question, if you ask it the way you did, is no, I can't be prepared to sit here today and say that I know the best immunosuppressive regimen or that there is preclinical data to suggest what that is.

DR. SALOMON: I guess I was avoiding that.

MR. SIEGEL: Can I please--let's try to look at the questions before you jump to conclusions about what we are asking and what advice we want. There is not a question here about is there a standard regimen, should there always be one drug.

The questions here are specifically focused on our regulatory needs. Each and every one of them is about islet-only therapy. Why is each and every one of them about islet-only therapy? Because islet-only therapy is exposing patients to immunosuppression who were not otherwise to be exposed to immunosuppression.

It is there where we have a significant burden to determine whether this is a reasonable and unnecessary risk. It is there where we must ask, do we yet know enough from animal models to do this? What information should we have from animal models? What would be the most appropriate patients from a potential benefit or rationale to risk to make that determination?

We are not here to say, "This is the right immunosuppressive regimen for islet transplants." That is not even the question on the table.

DR. SALOMON: Again, the question that I was posing to try and follow, really, the spirit of what Jeff said and what Jay said is based on the animal-model

experience that we discussed this morning and some yesterday, if you wanted to go forward into a clinical trial, which animal models or model would provide the kind of data to justify a given choice of immunosuppressive drug?

I am not trying to tell you to say that it is rapamycin; just any approach, what approach? If the answer is there is no approach, then we need to tell the FDA that which means, to me, that no one is ready to go to a clinical trial anytime soon, which is fine.

DR. AUCHINCLOSS: I beg your pardon? We are going into a clinical trial with the Edmonton protocol which was developed in the human animal model.

DR. SALOMON: The human animal model. So the idea, then, is that there is not preclinical animal--if that is what you want to say, that's fine. Then what Hugh is saying is that there is no preclinical data necessary. You choose an immunosuppressive regimen based on what? I am not certain. And then you start a human trial.

DR. RICORDI: Maybe I can rephrase Hugh's comment. I am saying there are many animal models valuable to develop new strategies and a research base for immunosuppression and to screen drug combinations and everything.

There is not a single animal model, in my opinion, that is a necessary prerequisite before moving to a pilot clinical trial because of the lack of existence of a model

similar to type-1 diabetes and that can predict safety or efficacy in human patients.

DR. CHAMPLIN: As I mentioned this morning, there are big problems in trying to take drug trials from animals directly into humans. Certainly, we have the data that was presented this morning. We also have human data. So I think that that is, perhaps, the most important data as one considers going forward as to where are we now.

At least from the inklings of what we have heard of the Edmonton protocol, it sounds to be a successful starting point.

DR. MILLER: I agree with Dick. The question we are sort of struggling with here, I think, is how to integrate what we already know about humans when you are now asking us to go back to the animal models.

So a question that may help is do we feel that we can take a pilot trial without any further animal data and generalize it and therefore leave the next steps to what animal models you need to do before you then go back into a different protocol for a human trial.

I am not exactly sure of the number of patients treated with islet-only cells in the pilot trial, the preliminary data, to then going on and building this multicenter trial. We don't know how strong the data is even though we hear it.

So my feeling is that there is enough clinical data already that can be reproduced or that could be validated and looks good that shows that they followed so many people out so long. You are not going to get any better than that for the study of this current regimen.

Therefore, if it is valid and reviewed, that probably is enough to go ahead and do the pilot trial. If that is our first answer, then the second question is, okay, now we want to get away with no immunosuppression and what kind of animal models do you need to do that. That is a separate, question.

I think it is a separate question than whether or not the pilot trial is adequately controlled by the human experience we already have.

DR. BLACK: Could I take one step back and say just surely the team, Dr. Shapiro and Dr. Lakey, here, did not arrive at their FK506-rapamycin combination without preliminary work, perhaps dating back a number of years or in several different models that gave them suggestions of how to proceed in the clinic.

If you could clarify that a little bit?

DR. SHAPIRO: That is exactly right. Our trial is built on a synthesis of many years of preclinical experimentation and also a substantial clinical knowledge of use of these agents in different usage and combinations in

kidney transplantation and other models.

We haven't just come to the scene here with a brand-new therapy that hasn't been tested or applied in other situations. We have just used a cocktail of agents that we believe are safe to use clinically in a different way.

DR. SALOMON: What models in that development process did you think gave you the best information or were they all just pieces of a complicated puzzle?

DR. SHAPIRO: I think it is like a jigsaw. Our preclinical in Edmonton had always been the adult islet-transplantation model. We knew that many of the drugs we use are not compatible with adults and we knew that that would only provide us information in terms of function of islets and provide us a little bit of information in terms of toxicity but not sufficient.

And then you synthesize also what is available clinically and what has occurred in that realm.

DR. SALOMON: So, Dr. Kenyon, in that regard, where does the non-human primate studies that you are doing--they are very expensive. They are very involving. If this is just a complex jigsaw, maybe we could save some serious money.

DR. KENYON: It is a serious effort. No; I agree with everything that they have said so far, but, clearly. I

would not be comfortable going into a human with an agent, for example, a new monoclonal or a new agent that had no experience in the clinic.

So I think that is where the non-human primate models, the dog models, the animal models--we get our suggestions of efficacy from the rodents and then we have to move them up and see if they work in the larger animals.

I think that is very important there, but, clearly, that doesn't give you the final answer either, so I am not going to sit here and say that is the final answer. It is clearly not true, but very important for new immunomodulatory agents.

DR. AUCHINCLOSS: We need to be clear, again. I am not arguing that new immunoregulatory agents should not be regulated by the FDA. They are and they should be. So the FDA will certainly regulate a trial that had, for example, anti-CD154. That is not the question.

The question is whether they are regulating islet transplantation, themselves.

MR. SIEGEL: These are unapproved uses of all these drugs. Even if there was no cellular component, this trial would require FDA review. The rapamycin, the FK506, they are not approved for this use.

DR. KENYON: Hugh, I think they are really asking where do you draw the line? Clearly, in the type of study

they are doing in Edmonton, there is enough clinical data plus their experience to support it. So I really thought that what you were asking is where do you think it is important to have some preclinical data.

I think, clearly, it is where you don't a lot of clinical data. But I, personally, would not want to do a human transplant with an agent that had really never been used clinically without having some preclinical data. I think we do.

DR. BLUESTONE: I think what we don't agree on and what we haven't come to closure on--and that is why I don't like the question. What the question is posing, in a way, is that there is a paradigm model out here, that there is some set of three models, if you put them together you should be able to--so my answer to the question is that, in the Edmonton case, then Hugh is right. But in the CD154 case, then Hugh wouldn't be right because there isn't a human experience to rely on.

And he does not disagree with me. So the answer, from my perspective, goes back to what Jay says, you have to make a rationale argument. How do you make a rationale argument. It has got to be a combination of preclinical and/or clinical experience that demonstrates safety and some degree of efficacy.

How you actually build that equation up is the

same jigsaw puzzle that Jim has already done. It is hard to sit here and say that an animal model, or two animal models, are going to be the answer or not. You have to base the recommendations on a series of identifiable results that make it a compelling rationale to go forward.

I don't think it is always going to be due to monkeys, and it is not always going to be a NOD mouse. But sometimes a NOD mouse might work because the antibody didn't work in a monkey. And sometimes the monkey will be workable because it does work.

I think it would be a mistake to try to set a clear set of parameters of what those preclinical trials should be other than the general principle that, in the absence of clinical data, compelling clinical data, you need preclinical data.

DR. SALOMON: I don't disagree. That is kind of what I was trying to get at when I used the analogy in talking to Jim about the fact that it is a jigsaw. So I think, so far, the message that I am very comfortable in giving to the FDA is that preclinical models are going to be critical, but that there is no single preclinical model that will give you an answer or that you should require information in.

It rather should be a presentation of a logical series of preclinical experiments. I think that also, and

again I would put this out for discussion, it would be equally wrong that if someone came with a single preclinical model and you wanted, based on a single preclinical model, really without much evidence surrounding it, to go forward and do a clinical trial, that probably would be equally questionable, that there should be, probably, in any situation, a series of models, a series of lines of investigation, hopefully independent, all of which rationally support the decision made for the clinical project.

Do you agree with that or not?

DR. SHERWIN: I do. The only thing that I would add is that, particularly with this disease being an autoimmune disease, that one should consider the possibility of using an autoimmune model of diabetes within that mix because there is a certain level of complexity to the problem.

It is not a problem when you use big drugs. The drugs you used, it wouldn't matter at all. But when you start getting to refined--because we are talking about people without kidney transplant. That is a different story. Consequently, the kind of immunosuppression that you use in that setting should be less toxic, I think, than the kind of immunosuppression you would use with a kidney graft as well.

So when you get to the more selective agents where you are not going to be hitting it with a sledgehammer, I think, at that point, an autoimmune model could give you answers that are important and I wouldn't ignore that.

DR. BLUESTONE: Would you say that the drug should be less toxic than if it was a pancreas transplant?

DR. SHERWIN: I am not too in favor of pancreas transplants in people who have no significant or serious complications. In other words, we use pancreas grafts, but, usually, it is in people that are either having impending renal failure or require a renal graft. So it is not a routine thing, I think, to use pancreas transplantation in people--see; we disagree, obviously.

DR. HERING: Let me ask you. You are a diabetologist.

DR. SHERWIN: Yes.

DR. HERING: Would you admit that patients with hypoglycemia unawareness and defective glucose-count irregulation, would you consider this a complication of diabetes?

DR. SHERWIN: Of course it is. But with good care, generally speaking, that can be dealt with.

DR. HERING: With good care in a clinical research center; right?

DR. SHERWIN: No, no, no, no. You know, there are

people that have recurrent hypoglycemia where it is a problem. But, first of all, we have much better systems and we are evolving much better systems for monitoring. Consequently, in the next few years, those problems are going to diminish.

So I rarely encounter somebody that I would think--there are, like, one or two patients I have encountered in the last ten years that I would think about it. There is a rare patient where you are correct. But I disagree. I think with more effective insulin delivery systems and good management, most people can be managed without enough of a problem to warrant immunosuppression.

I would rather have recurrent hypoglycemia periodically that is manageable than immunosuppression.

DR. HERING: I guess the DCCT was published in '93, yet diabetes remains one of the leading causes of blindness and this and this and this, well, at least in the type-1 diabetic population. Would you accept that intensified insulin treatment is difficult to implement?

DR. SHERWIN: Of course. Of course it is difficult to implement, but the improvements that one achieves with intensified therapy are sufficient, I think, to warrant real caution doing a pancreas graft to try to take it to the next level because the risks of imposing that are greater than the risk of the disease, I think.

We are getting into the wrong discussion, I know. Maybe we should stop.

DR. CARA: Actually, I think this is just the right sort of discussion to have because I think we sort of took the cart before the horse when we started talking about different sorts of regimens in the sense that it seemed that we had all sort of accepted the notion that islet-cell transplantation in a patient without any evidence--or on a patient who is not already on immunosuppression therapy should continue forward.

I am not sure that we all agree with that. So I think the issue that we really need to sort of look at is whether or not there is enough information, there is enough data, to carry forward with islet-cell transplantation in patients that are not on any sort of immunosuppressive therapy.

MR. SIEGEL: This is the question that is, I think, the first point in the question--

DR. SALOMON: We haven't forgotten that one, Jay.

MR. SIEGEL: The first bullet is specifically about which patients for the very reason of the nature of what the alternative treatments are that are that available and what is the course of the disease--this is an important one in our construct in determining what are acceptable risks or acceptable--

DR. SALOMON: I would point out that actually what Bernhard and Bob were doing was discussing this question. So I don't want to derail that now because that is--and I agree with Dr. Cara that this is what we ought to ask now, and that is what is the population of patients, if any, that you believe today would be candidates for islet transplantation only, not in the setting of a kidney or a liver.

DR. SHAPIRO: Patients essentially who have documented evidence of failure of exogenous insulin therapy for whatever reason.

DR. SALOMON: Can you help us decide, what would that be? Recurrent malignant hypoglycemia? There has been discussion in the literature on what that population should be. I think the FDA wants to hear us discuss what those patients might be.

DR. SHAPIRO: I think those opinions vary between expert diabetologists. It is very difficult to define. The patient that we say has totally failed all efforts at optimal glycemic control despite very intensive insulin therapy, Dr. Sherwin would say he could easily treat that patient.

DR. SHERWIN: I am not trying to say that. I am not glib. I have as much trouble as anyone else. I think that the improvements that one can achieve with optimized therapy--there are now glucose sensors that are about to

released that can give more continuous monitoring of glucose which is going to provide a lot of information.

DR. HERING: I agree 100 percent. Eventually, we will have to perform a prospective clinical trial to address this question. Now we have to identify a patient population to get islet transplantation to the level--

DR. SHERWIN: I would say in people with incipient creatinines, let's say, above two and a half or three who still have function, to me, that would be not an unreasonable group of people.

But I think you would have to be very, very careful about using hypoglycemia as a dominant reason to do an islet graft. It may be that there are selected individuals who do that, but I would suggest that it would require some sort of independent team of people to assess what had been done clinically prior to subjecting someone, on that basis, for allotransplantation.

So I think you would need an independent team of, perhaps, experts to evaluate the situation and be sure that medical therapy had been exhausted. I am not saying that there aren't individuals like that, but there are not many.

DR. HERING: A fundamental point here is we are not recommending islet transplantation for the treatment of hypoglycemia unawareness. But we are going to identify a patient population and we want to see whether, in a

controlled clinical trial, we can test whether islet transplantation can be performed safely and in an effective manner.

So we are not recommending this as a treatment. We are identifying a population that is considered suitable.

DR. SALOMON: Can you be specific about what that patient population is, then, in the trial that you are proposing?

DR. HERING: Also, we can ask what are the recommendations of the American Diabetes Association for solitary pancreas transplantation. ADA has concluded that it was "patients in whom all other measures have consistently failed to ameliorate the situation;" this was the definition.

DR. SHERWIN: That's true. It depends on how you judge that. So I am saying that it really would require, I think, some sort of assessment by an independent team of diabetologists to say that it had been exhausted. Then I think it is acceptable.

DR. HERING: I think that is a wonderful agreement. I guess nobody would argue that a diabetologist should be part of the team and should be approached and should probably refer the patient.

DR. SHERWIN: My only concern is that it really be an independent sort of advisory committee or something like

that because it is not a problem when you are dealing with people with renal disease who are going to require a kidney graft. If you don't want to have people on immunosuppression, you could study people who one could predict within a short period of time, they will require a kidney graft even though they don't require immunosuppression--

DR. BLUESTONE: Isn't that the job of the IRB?

DR. SHERWIN: I would be careful about IRBs because they don't treat people with diabetes. They are not sophisticated in the ways of patient selection. I think IRBs can evaluate certain issues of importance but I don't think, in really exposing people to initial trials where we really don't know too much about outcomes yet, to me, if you really want to avoid immunosuppression in a model that has immunosuppression, you should focus on people who don't have it now, who are about to need it.

DR. MILLER: But isn't it the role of clinical trials and the informed-consent process for the physician taking care of the patient to discuss the risks and benefits? You have to have some minimum amount of disease in order to say, "This is a bad group." But I don't think you have to have every patient determined by a group to say that, yes, this patient has failed everything else, because that is part--each person's risk/benefit ratio participating

in the clinical trial is different.

So I think you have to set standards saying what is a minimum level of severe diabetes to make it, but I think that as long as it is being done in the context of a well-designed clinical trial where the patients are informed of the risks and benefits, and they have a disease which could potentially be benefitted by the intervention, that that is what informed consent is all about.

DR. SALOMON: Can you guys help me? I guess that question that I am thinking about is we have the possibility of--we are doing kidney transplants, obviously, in patients with diabetes. Then the idea is that we have done a lot of islet transplants and pancreas transplants in that group.

Now, and correct me if I am wrong, but you have made a decision that you want to do the next group as islet alone. I am not objecting to that in any way. Can you help us with how you--

DR. HERING: I think the point to make is the following. Islet transplantation has been performed in settings of simultaneous previous kidney or liver transplantation. This was probably okay to study some of the basic questions, but at the very same time, I guess progress was slowed by the fact that you are doing transplants only in patients that have received a kidney transplant because now the kidney determines the protocol.

This is not the way to proceed. There are very well-defined issues in islet transplantation such as whether the treatment is diabetogenic, whether the treatment would control autoimmunity, that are completely irrelevant, more or less, in kidney transplantation.

So I think, for this specific reason, we think the field can only progress if you can address the questions that matter without considering all the very important issues in kidney transplantation.

DR. SALOMON: I think that is well said, Bernhard. So, can we begin to just analyze what is that population, then? I have heard general statements like they should be bad diabetics, I guess, is as close as I can get so far.

MR. SIEGEL: Let me categorize what I have heard because I am confused by this dialogue, and maybe I missed something. I thought there was general agreement among the speakers that that population is "failure of exogenous insulin therapy."

What I heard Dr. Sherwin say, if I may characterize, is that if one of the indicators of failure is recurrent hypoglycemia or undetected unsymptomatic hypoglycemia, that one needs to be particularly cautious to insure that that truly is failure of insulin therapy, that that patient has been appropriately treated with the state of the art before exposing him to these risks by appropriate

independent experts.

I don't think I heard disagreement. I heard some discussion of informed consent or IRB issues, but not disagreement, that one should insure that that is the case for those patients.

DR. LEVITSKY: I actually heard the other day another category of patients which I commented on then and would like to get back to again which was the patients who were failures because they had ketoacidosis.

I am very concerned about including that group. As a matter of fact, both groups are of concern to me because there is not only a biologic base for these disorders but also a psychological base. If you have a group of patients who have not been very carefully screened and not by a diabetologist who is fully committed to your project and your patient and on the payroll, I am a little worried that you will actually be collecting a group of patients who are going to be much less adherent to your immunosuppressive regimen than if you went, for instance, to the group which Bob Sherwin was discussing which is a group which is about to go into renal failure but isn't yet and so, therefore, we can predict that decline in renal function and when it is going to happen pretty well, even with ace inhibitors and whatever.

So that population would tend to be a population

which would offer you, I think, more stability from the psychological point of view, perhaps, than the group that I heard selected out before.

DR. SHAPIRO: As I have said yesterday, transplanted patients who specifically have only metabolic instability or only ketoacidosis--these are patients who have real failure despite tremendous efforts on their part independent evaluate to confirm that, independent psychological evaluation in selected cases where appropriate, to confirm that they really are failing on the best management that we are aware of today.

DR. LEVITSKY: Failing with ketoacidosis as well as hypoglycemia?

DR. SHAPIRO: In certain cases, occasionally; yes. We are talking about a very highly--

DR. LEVITSKY: I would propose to you that any case that fails with ketoacidosis as well as hypoglycemia doesn't have a biologic problem. I cannot have that biologic problem defined. Maybe Dr. Sherwin can define that, but I cannot.

DR. BLUESTONE: I am a little confused. There are two things here; right? There is the islet transplant and the immunosuppression. It sounds to me like I have not heard anybody think that that safety issues surrounding the islets, themselves, is an issue to be concerned about, that

the issues have been focused on immunosuppression.

That is what I heard. So, if that is true, you would ask, what about if the patient doesn't get an islet transplant. The reality is that there have been and are ongoing clinical trials in new-onset diabetics who have none of these very severe things with immunosuppressive drugs including cyclosporine which has been tested and things like that in a patient population which I would imagine, on the face of it, you would say is not a patient population you would be submitting to these immunosuppressive therapies.

It has nothing to do with islet transplantation. It has to do with whether or not we think it reasonable in diabetics, given some of the morbidities and the outcomes, of trying to test novel immunosuppressive therapies in those patients, with or without islets.

So if the issue is about immunosuppression and not about islet transplantation, then I think it takes it into a whole different realm of how do we treat our diabetics with regards to immunosuppression.

DR. SALOMON: The fact is that I was at the NIH at a meeting with Joan. It was in about 1998, where the discussion was with the experts, pediatric diabetologists, transplant people. I was there representing immunosuppression.

The message we got pretty darned clearly was that

we were not going to be allowed to do any sort of major trials of immunosuppression in these diabetics.

DR. BLUESTONE: They didn't listen to you.

DR. SALOMON: No, no; they did listen to us. Most of those trials were over.

DR. BLUESTONE: Since then, there have been steroid trials. There have been cyclosporine trials. I was at a meeting--maybe it was the same meeting.

DR. SHERWIN: They were short-term trials.

DR. BLUESTONE: Well, it was up to a year. These may be short-term because if the islets don't take--let's think about an outcome here.

DR. SHERWIN: But this is a long-term trial. I hope it is.

DR. BLUESTONE: Right. So the question is that if you get normal glycemia after the islet transplant, and a year out, you are asking the question--a year out, with a normal glycemic patient with a functioning transplant, would you be more worried about keeping them on immunosuppression at that point or taking them off because of the long-term immunosuppression?

You would keep them on immunosuppression because what you have gained from making them normal glycemic outweighs the risk of the long-term immunosuppression or that point, or would you take them off because you would

rather have them lose the graft than have long-term immunosuppression?

DR. SHERWIN: I don't know.

DR. SALOMON: I would just point out that I was there in 1998 to convince them to do the immunosuppression, Jeff, so it is not always clear what agendas we had at different times. I am not against immunosuppression now. I think, though, we are getting away from what I wanted to do to finish this first topic which was at least finish discussing what would be the candidate for this first trial.

I think the comments of Bernhard and James have been on the point there. One population that I would like to ask you guys about would be relatively young patients which, of course, means within five years of my current age, who have microalbuminuria. You get the idea; some sort of microvascular disease that was easily objectifiable and yet were certainly far from the serious downstream complications of diabetes.

What about that as a population that could be used for these early studies?

DR. SHAPIRO: Clearly, that is an important population group for studying the long-term efficacy of islet transplantation is the control of secondary complications. It is not the population that we are targeting in our multicenter trial right now partly because

we are a little concerned about the use of the low-dose tacrolimus and its potential tacrolimus effect.

The last thing we want to do in that patient population is accelerate the nephrotoxicity.

DR. RICORDI: Actually, I think to evaluate the patient population is very important to establish the baseline of what is the standard of treatment. The most worrisome thing that I heard today from the diabetologists is that we are happy with what insulin can do today and that hypoglycemia unawareness is not a real clinical problem.

DR. SHERWIN: No; that is not what I said.

DR. RICORDI: No, no; I heard very well that you said that it can be treated. Incidentally, I want to put on the record the reason next to my name there is Stacy Joy Goodman Professor is that Stacy Joy, a sixteen-year old, died in a hypoglycemia crisis and this is for sure an element that can threaten the life of patients.

Maybe it can be managed at highly specialized institutions or maybe the standard of care will improve the life of patients with diabetes, but so far the gold standard of insulin treatment through intensive-care treatment and normalization of hemoglobin A1c through intensive insulin treatment can be achieved in less than 5 percent of patients with diabetes.

That is why I believe there is uniform agreement

that insulin has been ineffective to prevent the complications of the disease that can develop and the reason why we spend $120 billion a year on diabetes and it is one of the leading causes of death, amputation, blindness and kidney failure.

Otherwise, we would not be here. I don't have the representatives from JDF or the parents of these children who died or who have this complication, but I personally consider it outrageous that since we can treat and control everyone with insulin, why assume any risk.

These are calculated risks that have to be put forward to move a field of critical importance forward.

DR. SHERWIN: I am not that naive. Believe me. I think that the issue is to take a step-wise approach in a new form of therapy. I want this to work and I really want this to work. It is not that I don't want it to work and it is not that I think that insulin therapy that we currently offer is optimal.

But I do think that, in a situation where we don't know outcomes yet, even though we think we may know from short-term experiments, we don't know long-term results. we don't have all the information and it is best to start in small steps.

DR. RICORDI: We started with six patients and we are now going to 28 in a very controlled--

DR. SHERWIN: Well, six patients for how long? See, I don't have the data. But how long?

DR. RICORDI: We are not talking about a vaccination of all children.

DR. SHERWIN: No. What I am saying is somebody, two years from now, may get cryptococcus or somebody else--

DR. SHAPIRO: The precise risks of what we are proposing to do I think will come out with a very carefully conducted prospective clinical trial.

DR. SHERWIN: All I am saying is, when you start a trial, generally speaking, you would like to have very well-defined criteria. When you start to get into the subjectivity of how much hypoglycemia is a problem or how much ketoacidosis is a problem, that becomes a very subjective assessment and is based upon lots of factors.

A certain level of creatinine or a certain level of proteinuria is a nice hard endpoint that allows you to begin to approach a problem in a patient population that it is a very high risk.

It seems to me that that is the population in these early stages--it is not that I don't want everybody to get islet transplants ultimately. I think it is the way to go over the long haul, but I do think, in the early stages, it is a potential risk because our therapies are primitive, still.

We have so much to learn. We don't know everything at this point. I just think the highest-risk people that is not going to screw you up in terms of assessing islets only would be the optimal patient to study. That is not a subjective assessment.

DR. AUCHINCLOSS: I think I disagree, essentially, with everybody here except for Carole because I think the only criteria that is important here is informed consent. In my view, if I had had diabetes for fifteen years and I had zero complications of diabetes and I was under perfect control, from the data that I have seen from Edmonton, I would take the choice to enter that trial because I would see it as a no-lose situation.

I either get an islet transplant working and I am on, I think, a very non-toxic program of immunosuppression or it fails and I stop the immunosuppression. I think it is an entirely justified trial for every patient with diabetes, maybe with a minimum time period that they have had it as long as you have informed consent.

DR. HARLAN: My name is David Harlan. I am from the NIH. I was going to make the point that Hugh made but also to add the point that if you wait until a person has incipient renal failure or significant proteinuria, then you run into a problem where we know that they are likely to require a kidney transplant and that pancreas-kidney

transplant is known to be very curative, 90 percent curative.

Then you are depriving them of a known benefit in that population where an islet transplant alone is not known to be as effective. It is 90 percent cure rate if you have got a kidney-transplant recipient. We don't know that with islet.

DR. SHERWIN: I thought the question was how do we begin trials in people who are not getting kidney grafts? That was the question. What I am saying is that you are absolutely right, that it is not going to be terribly effective in terms of preventing them from going into kidney failure down the road. I will accept that. Is that what you are saying?

DR. HARLAN: My main point is the one that Hugh Auchincloss made.

DR. SHERWIN: So you would take anybody who came to you off the street--

DR. HARLAN: With brittle diabetes--

DR. SHERWIN: And has informed consent.

DR. HARLAN: If it is truly informed consent. We can argue about what is informed consent because that is a difficult thing to truly achieve.

DR. SHERWIN: Do you think we are that far along at this point?