FOOD AND DRUG ADMINISTRATION

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C O N T E N T S

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P R O C E E D I N G S

Welcome and Opening Remarks

DR. JONES: Good morning. I was just sitting here writing my remarks. They sent me out of town without anything, but that is okay. Sometimes it is better that way.

I am Deputy Assistant Secretary for Health for Women's Health In the U.S. Department of Health and Human Services. Our office, the Office on Women's Health within the Office of the Secretary-- now that I have said all that, my time is up--is quite pleased to be a cosponsor of the meeting this morning.

It occurred to me--despite all the churning and thinking about what we were doing and what we hoped to accomplish in this workshop, it occurred to me, as I struggled to get here from three miles down the road and it took me an hour and fifteen minutes this morning, that my journey was not unlike the ART journey in that there are two sides.

There is, if you will, an industry or a provider side and there is a consumer side. There is a goal to get

somewhere or to, if you will, produce an infant for a couple highly desiring a child. There is a lot of work that goes on behind the scenes unknown by each side and, perhaps, appreciated and still largely a mystery.

I have no idea what the Metro system does to keep it running on time and on schedule, but I expect it to be there when I go wanting the safety. So that is not unlike the consumer, the couple, the woman who wants a child who comes for ART technologies and expects the technologies to be there when she needs them, to be effective and to yield the expected outcome.

Today and tomorrow, we are going to focus on the science, on the animal models and some of the ethical issues underlying the techniques and processes. It is a terribly exciting time because this entire technology has come so far in the last quarter century, and there is still so far to go. But there is so much it has taught us, so much it has yielded, that this may well be the best time to be undertaking the discussions that we are about to have here.

So I welcome all of you. I look forward to a very exciting day and a half and look forward to continuing

to work with my colleagues from NIH and FDA as well as those from CDC with whom we have worked over the past three or four years in an Assisted Reproductive Technologies Working Group within the Department, just sharing information and keeping ourselves up to date on each other's activities.

So I wish you a successful day-and-a-half meeting. I expect to be able to spend the better part of here with you learning something, myself, because I do so seldom get to do anything science anymore. It is easy for me to be here rather than be downtown. It is an easy choice.

So thank you all very much.

DR. ALEXANDER: Good morning. I am Duane Alexander. I am the Director of the National Institute of Child Health and Human Development which is one of the co-sponsors of this meeting. It is my pleasure to welcome you and thank you for taking time to join us for this very important and useful meeting.

We expect, from the federal side, to learn a lot from you in this meeting and I hope, from your side, you learn a lot from your participation as well.

The NICHD has been in a very special situation over the course of the years, really since 1973 but, particularly, since 1980, with regard to research support for assisted reproductive technology. We have been under restrictions in our ability to fund human research in this area but with wide ranging ability to fund any nonhuman research.

The NICHD has taken the position that, then, we would push the limits of what we were to be able to learn from assisted reproductive technology research and animal models. Examples of this success have been the Culture Club, as it was popularly called, that Dick Cass organized and which has contributed enormously to fundamental knowledge of reproductive processes and ways we have learned from animal studies methods to improve the culture and development of fertilized eggs as they grown into embryos and improve the success of in vitro fertilization.

We have also studied other assisted reproductive technology processes other than IDF where the restrictions have not been so intense. We have also supported work in primate models such as Gerry Schatten's that you will hear about, and so we have done, I think the best would could to

push the limits of what we were able to do with the restrictions that were being placed on this area of research.

You will hear today from a number of presenters about what has been learned and accomplished from that from Dr. Biggers, from Dr. Schatten and others, Dr. Guidice, and the work that has been accomplished in the face of what we were limited in doing

So I think we have contributed significantly but there is a lot yet to learn. There is also the opportunity to do more on the human side than we have done in the past. I think, in many ways, investigators have labored under impressions that restrictions were more stringent than they actually are. Also, we were restricted in areas across the board in assisted reproductive technology rather than just in vitro fertilization.

So I hope that this conference will also provide an opportunity for clarification of just what some of these restrictions are and what opportunities may exist that are beyond what people may believe at the present time.

So we welcome your attendance. We hope that you will learn. We expect to learn from you. I wish you the very best for a very successful conference.

Thanks.

DR. ZOON: Thank you. My name is Kathy Zoon. I am the Director of the Center for Biologics Evaluation and Research with the Food and Drug Administration. I take a particular pride in helping co-sponsor this important meeting. Many reasons for this. Both Duane and Wanda have pointed out our efforts over the past several years, really trying to formulate a Health and Human Services position in this area and especially, as we go forward in looking into the future of ART, how we are going to develop a regulatory framework that will help assure the quality and safety of these procedures as they go forward.

So today's and tomorrow's discussions of inputting into the science of ART is going to be extremely important and, to us, we are going to both listen and present in a way to exchange information. In working with our colleagues, we recognize that, as part of the government, it is important for us to work with all interested parties, and societies and professionals in this

area, so we have a good understanding of what the current environment is because, in terms of looking at the science and making sure that it goes forward in an appropriate way, we don't want to be overrestrictive or underrestrictive in developing a regulatory framework.

It really is important for us to base our regulatory framework on good science. Really focussing on the science over these two days, I think, is clear it is not the beginning of our process and it is not the end of our process. We are in the middle of our process. We were collecting information and working on the science and trying to develop the questions that we think are important to answer with good science as we move forward into this area.

Right now, the Center for Biologics has recently just announced our new Office of Cells, Tissues and Gene Therapies. I think this is really important because many of the initiatives with tissues, with reproductive tissues, ART, will be under auspices, in FDA, under this new issues. We work very closely with NIH, CDC, the Office of Women's Health, both at the department level and at our own agency

level, who have been very supportive in moving this program forward.

My own sense is that we will listen very carefully over the next two days and work with all of you in trying to understand what are the issues that still need to be addressed and what are the best scientific approaches that we can use to address these questions as we move forward.

I wish you all the best. I think it is a great program. I think it should really stimulate a lot of very positive and useful discussion. I would also just, finally, in closing, thank the organizers for all their help in putting this meeting together and I wish you the best success.

Thank you very much.

FDA--Brief Outline of Regulatory Framework for Human Cells and Tissues.

MS. WARNER: Good morning.

[Slide.]

I am Jill Warner. I work with Kathy in the Center for Biologics, Evaluation and Research. I am going to just give you a little overview of our regulatory

approach for human cells, tissues and cellular and tissue-based products which includes reproductive tissue, give you a sense of where we are right now in the development and some directions that are open to sort of learning from this experience and others in terms of how we might develop the approach towards reproductive tissue and manipulated reproductive tissue that is commonly used in the ART procedures.

[Slide.]

We rely on three basic statutory authorities for the development of our regulatory approach. The Public Health Service Act, Section 351, gives authority to license biological products. The Food, Drug and Cosmetic Act is authority for the regulation of drugs which include biological products and devices, medical devices.

Finally, Section 361 of the Public Health Service is authority to promulgate regulations to control the spread of communicable disease.

[Slide.]

In 1997, we announced a new proposed approach to the regulation of human cellular and tissue-based products. This was selected as a Reinventing Government initiative.

The scope of this approach is very broad to include a wide range of human cellular and tissue products because we have found that, in the past, the regulation was somewhat fragmented and we wanted to put into place a tiered, risk-based system that would address a wide range of products in a consistent way.

Most of this approach is going to be implemented through rulemaking but, as I will note later, we are relying on existing authorities and regulations also for some aspects of the regulation.

[Slide.]

It might be helpful to sort of simplify this in terms of there are essentially two basic tiers of regulation. The first tier includes all those products that we will be regulating solely under these communicable-disease regulations.

The regulatory approach really looks at the characteristics of the human cellular-tissue product. In terms of it asks do we have certain characteristics that raise more significant clinical safety and effectiveness issues or are these issues that can basically be dealt with

by donor screening, donor testing, good-tissue practice, which I will tell you a little bit more about later.

But the elements of the types of tissue that will be regulated under the first tier; if the tissue is minimally manipulated, not highly manufactured, not highly changed in its sort of genetic or cellular characteristics; if it is intended for homologous use--that essentially means it is going to be used for the same purposes that it served in the native body; if it is not combined with a drug or device, it is not a tissue-engineered product or a combination product that includes a device component; and it doesn't have a systemic effect.

Now, for reproductive tissue, we recognize that reproductive tissue has a systemic effect, yet we still regulate it solely under Section 361 authority if it has these other characteristics.

[Slide.]

The second tier are those products that do not meet the criteria. They may be highly manipulated. They may have other characteristics. These raise more significant clinical safety and effectiveness concerns. In

our proposed approach, we assert that we will regulate these as drugs, devices or biological products.

Because this is a tiered risk-based system, and all these products are derived from the human body and carry the potential for spreading communicable disease, they all will follow the communicable-disease requirements as well.

[Slide.]

So what does this mean for reproductive tissue? Basically, reproductive tissue, again, we are going to have those baseline requirements that we are putting into place through rulemaking that will help prevent the spread of communicable disease because reproductive tissue, like other human cells, tissues, carries the threat of spreading communicable disease.

The elements of these new rules will require registration of establishments with FDA, donor-screening and testing requirements, good-tissue practice. These are the sorts of controls on your manufacturing that assure that you have got the record keeping, that you have set up your processes and validated them.

Generally, good-tissue practices are analogous to our good-manufacturing practice but with a focus on communicable-disease prevention.

Labeling controls; the regulations would also provide for FDA inspection and enforcement authority in case of a product that is violative of the regulations.

[Slide.]

What kinds of reproductive tissue would require greater or more a more risk-based authority to regulate? Essentially, for reproductive tissue, we are talking about that which is more than minimally manipulated. Here, again, we are deriving this authority from Section 351 of the PHS Act and therefore are not relying on our rulemaking to obtain this authority. These would be regulated as biological products with a focus on demonstrating safety and effectiveness.

Just a place marker here. We are in the process, in this meeting and in other forums, of trying to get a sense of where this line should be drawn. We have, and I will talk about this in just a couple of minutes, a couple of cases where we have seen ART procedures that are very analogous to genetic transfer, to cloning, that we have

regulated, certainly, in other areas beside ART. We have seen these as not different.

I think we recognize that reproductive tissue is a unique area with unique issues. Part of the purpose of this meeting is to start to understand, as we go along, some of the areas where we might need to make adjustments in policy and regulations as we go along. There is opportunity for doing that in our regulations and our statutory authority.

[Slide.]

So the status of our rulemaking right now, and this is the communicable-disease regulations; we have issued a final rule in January of 2001 that requires registration and listing of the human cellular and tissue-based products. It will be effective for reproductive tissues when we finalize the other rules. So, at this point, it is not effective for reproductive tissues.

But this rule also sets out the framework in detail on how we will be regulating under the risk-based approached. The proposed rule for GPs was published in January, 2001. We are working on finalizing that rule and the proposed rule on suitability determination. This is

the donor testing and screening. It was published in September of 1999 and we are completing the final rule on that as well.

[Slide.]

So, as I mentioned earlier, we have recognized that some of the activities that are going on or that some individuals had started fall within our existing authority to regulate biological products and drugs. We did send a letter to researchers who were working in the cloning area to remind them that they did need to come into FDA for investigational new drug application if they wished to continue this activity.

However, this is sort of a unique case because we had decided, at the outset, that there were major unresolved safety issues and that an IND would not be appropriate to go forward at this time. We had sent a similar letter to investigational review boards back in October of 1998.

[Slide.]

Genetic transfer in July of 2001, we sent a letter to sponsors and researchers who had some indications that they might be using some techniques to transfer

genetic material from one individual to another by use of this ooplasm transfer or some other methods. This is very similar to gene therapies and gene transfers that we have regulated in FDA for quite a while and we felt that this needed to be handled in a similar way with oversight over the clinical investigations. So, again, INDs would be required in this case.

[Slide.]

A third example of where we have decided that investigational new drug applications should be used is where there is co-culture with nonhuman animal cells. This raises some xenotransplantation issues because the live animal cells can carry the potential for transmitting disease to the cells and then subsequently to the mother or offspring. Again, this is a risk that we felt should be studied carefully under investigational new drug applications.

[Slide.]

So, what is an investigational new drug application? Right now, we have regulations at 21 CFR Part 312 and they apply to clinical investigations of all drugs

and biological products, all those things that we are regulating as drugs and biological products.

These are submitted to FDA before clinical investigations begin and 30 days must elapse before you can start clinical studies. So if you don't hear anything back from the FDA, you can go ahead. Usually, there is some dialogue that goes on. If we find that there are unresolved safety issues, we do put a hold on that until those issues can be resolved.

[Slide.]

The principles behind an IND are to assure the safety and rights of the patient. We encourage innovation by allowing maximum flexibility in the early research. We assure the quality of the study design and this is primarily in Phases II and III, permit evaluation of effectiveness and safety.

Really, this is an approach that allows the researcher to focus in on those things that are working better, that are safer, that are more effective and, hopefully, lead to better choices that are available. We maximize the efficiency of the later application review by

promoting early consultation with the center and the agency.

[Slide.]

The amount and the type of information that is submitted depends on a number of characteristics; the novelty of the biological product, the extent to which it has been studied previously--this, again, is an area where animal data can help inform clinical studies, known or suspected risks and the developmental phase of the product--as I mentioned earlier, the Phase I is primarily for safety. Then, as you get into Phase II and III, you look more at effectiveness issues. And the scope and nature of the proposed protocols. The sponsor is the one who proposes the study design and so there is some back-and-forth through the FDA.

[Slide.]

The process includes some sponsor responsibilities. The sponsor is required to select the investigators and oversee their conduct, ensure compliance with the protocols and submit adverse-experience reports and annual reports.

[Slide.]

The investigator is required to ensure that the study goes along with the protocol, obtains informed consent from subjects and ensures investigational review-board approval and review.

One thing I should mention in the area of reproductive tissue, we do realize that there are some unique issues here. One of the things that we need to keep in mind as we develop policy and regulations in this area is that these are regulations and they can be modified and they can be written in a particularized manner for reproductive tissue. That is one of the things that we are keeping in mind as we go forward.

There may be specific areas that need to be modified because reproductive tissue is different than most drugs and biological products. There may be different informed-consent types of issues. There may be different safety or effectiveness types of issues.

[Slide.]

There are some additional resources for additional information. We have a website that gives as lot of information about our issue action plan and the

regulations and policies we are developing. Also, we have guidance for IRBs and clinical investigations.

Thanks very much.

NIH--Methods of Supporting Research in this Area

DR. TASCA: Good morning.

[Slide.]

I have been asked to come to speak this morning about the methods that NIH has used to support research in areas that are related to or directly involved in the assisted reproductive technologies.

First, I should say that the NICHD part, the National Institute or Child Health and Human Development, is the major supporter, the major sponsor, of research on all aspects of male and female health in the federal government. We support basic research. We support clinical research, clinical trials, behavioral studies and epidemiological studies.

[Slide.]

Within NICHD, the major support for the ART-related topics is the Center for Population Research. The Center for Population Research was started in 1968, so it has had a long history of supportive reproductive studies.

[Slide.]

Within the Center for Population Research, there are three branches. The Reproductive Sciences Branch--you can see I have highlighted that a little bit there, since that is the branch that I work in. That turns out to, again, be, both in terms of the volume or research and in terms of the amount of money spent on research on support of assisted reproductive technologies, the largest in the federal government.

The other two branches we have are the Contraception and Reproductive Health Branch and the Demographic and Behavioral Sciences Branch.

[Slide.]

In the Reproductive Sciences Branch, we have mission. A major mission is the alleviation of infertility. Another major mission is to develop novel contraceptive leads. As you all know in the audience, as I have looked at the list and I see that there are lots of people in here who have broad experience in reproduction, research that is initially targeted at alleviating infertility may give you leads that wind up being novel contraceptive leads and vice versa.

Within the Reproductive Sciences Branch, we support all aspects--cellular, molecular, genetic aspects--of reproductive endocrinology, reproductive neuroendocrinology. We support, again, cell, molecular, genetic aspects of gametogenesis, fertilization, pre-implantation development, implantation. We also support certain reproductive diseases such as the polycystic ovarian syndrome, endometriosis and other related diseases.

[Slide.]

How do we support these? Again, this is a long history and we are talking about many, many grants over thirty years of support in all different areas of reproductive biology, as I have mentioned.

We use the following funding mechanisms: investigator-initiated research projects--these are RO1s or regular research grants that people send in; program projects; small grants, which are called RO3 grants, as well as small-business grants. We also use mechanisms that involve solicitation types of grants. These are requests for applications or RFAs, PAs, program announcements, and also the request for proposals, contracts.

We have a large program of training, fellowships, career-development awards that are available for people who are interested, or those who are appropriate for getting funding. Through the years, I can't even begin to tell you how many conferences and workshops we have had, and in many of these we have made a very strong attempt to bring together clinicians and basic scientists in the same room and have tried to get them to speak together, to speak to us and give us some information about what are the needs in the field and maybe some ideas about how they think we might go forward with these.

[Slide.]

One of the things that we talked about here is human embryo-based technologies. These human embryo-based technologies--I have listed some of these at the bottom of the slide here; in vitro fertilization, ICSI, cytoplasmic transfer. I think you are pretty familiar with most of these; pre-implantation, genetic diagnosis, embryo culture.

A lot of things that we are unable fund. As Duane mentioned earlier, once the egg has been exposed to sperm, then we are not able to fund that. So those items that are on there are--we are able to do it, but we have to

use nonhuman organisms to do that. So that was the thrust of what we tried to do.

As you can see, there is one item on there that is in yellow so that we can fund, on this list of human embryo-based technologies, and that is the embryonic stem cells, from stem-cell lines that have already been established. But that is another story. I am not going to talk about that today.

How can we do this? How can we do this funding despite the limitation of not being able to fund work between the one-cell stage and the blastocyst stage of embryo development. I was going to tell you that the answer is plastics. That might work in certain places. I think maybe if you are of a certain vintage, maybe that works but what we have decided to do, and Duane alluded to this, in about roughly 1986, we had the idea that we could use a mechanism called cooperative agreements to fund research.

So our branch, from 1986 until now, has used, in addition to the investigator-initiated RO1s, which are really the bread and butter of the branch, we have used these cooperative-agreement mechanisms. Let me just tell

you a bit about that because that is on a slide I think that didn't get in here.

We issued an RFA in 1985, probably, which was called Nonhuman IVF in Preimplantation Development. This is what Duane referred to as the Culture Club. Now, it probably should have been called the Other Culture Club because there was another club. Again, that is if you are of a certain vintage, you may know what that was and if you are from a certain country, you may know what that was.

That was the Culture Club. That was designed, really, to use nonanimal models to try to improve the culture systems for preimplantation embryos and also for oocyte development in culture. The way that this cooperative agreements work is that we issue an RFA soliciting applications. The applications come in. The applications are reviewed and then we put together, we establish, a working group, a group of scientists who agree to collaborate with each other on, in this case, that particular topic of nonhuman IVF and preimplantation development.

I think some of the reasons--this group was extremely successful. I think some of the reasons for that

was that we were able to get together in one room a group of outstanding scientists who were committed to the project. They met together three times a year for sixteen years, it turned out to be. I don't think a single principal investigator missed a single meeting in all of that time.

So I think there was a lot of dedication and some of those people are in this audience right now. So I think that was a lot of the success of that, these cooperative agreements. The difference between cooperative agreements and grants and contracts is that a cooperative agreement is one in which the government has a substantial role.

In these cases, what happens is a member of the NIH staff is a partner in the research and serves as research coordinator or some other designation and with the role of trying to make sure that the research goes forward at a maximal rate of progress to try to keep things together so that we move towards the original goals of the program.

So that was our first cooperative agreement. These other items listed here are other RFAs that we issued. It turns out that all of these are directed at

ARTs in one way or another and some very heavily directed, targeted, like that Culture Club and also like the Trophoblast Maternal Interactions RFA. That one is to identify molecules that are involved in trophoblast and maternal interactions.

Upcoming is a Female Health and Egg Quality RFA that will start a year from now. All of these, except for the very first one on this list, are all done as cooperative agreements. Some of them are as described. The Culture Club is a group of maybe five, six, seven people who get together in a room and interact and collaborate.

Others are larger types of cooperative agreements. Those exist in the form--the Reproductive Medicine Network is one of those. The Specialized Cooperative Research Center is one of our major forms of support for research. This is done as a cooperative agreement. In this case, there are fourteen major medical centers involved in this and they have the main goal of supporting multidisciplinary interactions between clinicians and basic scientists. So this is, I think, a very important effort on the part of NICHD to promote

research related to the assisted- reproductive-technology technologies, that the specialized cooperative research centers are able to do a lot of things that can't really be done by individuals or by small groups of people but requires some fairly large networking.

They, for example, can establish research focus groups and have established research focus groups that cut across. They have representatives from several of the different institutions and they meet together on a separate basis and focus on a very particular targeted topic and in many, in probably all, cases is somehow related to the assisted reproductive technologies.

Then we also support some infertility research centers, also through a cooperative-agreement mechanism.

[Slide.]

I wanted to spend just one minute or two to tell you about the Culture Club since that was one of the good examples of how we could take this group of scientists and having agree to work together and show how some of the work that they produced has been able to filter over into the human IVF arena.

This started, as it says here, in 1986. Every five years or so, we compete so we would issue another RFA and have new applications come in so that the group gets replenished, renewed, refreshed. As a result, and I can only take a couple of minutes to do this, but if you look at some of these items that I have listed here, you will see that these are some of the items that have trickled down into the human IVF arena; the use of completely defined media, for example, to avoid the use of serum and other proteins that could bring pathogens or other harmful compounds into the culture media.

We promulgated, throughout the whole time of the Culture Club, the use of amino-acid supplementation. We discovered, in the mid-1990s, that inferior media could have an effect on genomic imprinting during the preimplantation period.

The development of embryos into blastocysts was a very important concept, not new with the Culture Club but very strongly promoted by the Culture Club and not just to be able to count the number of blastocysts but to be able to look at the blastocysts and say something about what the quality of the blastocysts happened to be, what are the

different ways of evaluating the quality of an individual blastocyst.

Barry Bavister, who is here in the audience, did a beautiful study published, what, around 1994, maybe, on the speed of cleavage of hamster embryos showing that speed of cleavage, the rapidity with which the embryo reached a certain stage, was directly correlated with the percent of blastocysts that could be produced and also with a doubling in the number of live offspring that could be produced. It was a very dramatic study. This, again, is something that is used today as part of the evaluation in human IVF clinics.

Finally, we had, as part of the Culture Club--John Eppig was constantly reminding us that it is really important to focus on egg quality, that the quality of the egg has a lot to do with what happens after that during embryo development and later development.

Other translational studies that have been supported by the Reproductive Sciences Branch of NICHD; we have a long history of support of biological history and chemical history of the gonadotropins and also supporting

the development of the recombinant gonadotropins that have been recently been put on the market.

We also have spent a lot of energy, a lot of money, a lot of time, on studies on the gonadotropin-releasing hormone and the analogues that are also used in the IVG clinics. Some of you may remember that, in 1977, Guillemin and Schalley got the Nobel Prize for determining the structure of GNRH. This was supported by the Center for Population Research in the Reproductive Sciences Branch.

We have provided strong support for research on protocols for ovulation induction, also for the follow up of ICSI offspring--this is mainly with monkeys and with mice--and also with predictors of IVF success. So these are some of the topics, I think, that we have covered.

It is impossible to tell you how much I think we have accomplished. Dr. Alexander mentioned this earlier, that we have spent, I think, a very large amount of money over all of these years. I think that we have been able to provide a lot of support, a lot of good solid research, in the area of reproductive sciences and in all of those areas

I mentioned before, especially reproductive endocrinology and all of the other ones that I mentioned earlier.

I would like to echo what he said, that I think there is a lot of room for applications to come in. We would like to receive applications. We would like to receive more investigator-initiated applications.

I think we will continue to utilize the cooperative-agreement mechanism. I think that has really been extremely useful in helping us to focus and target on the particular areas of alleviation of infertility.

DR. LEPPERD: I am Phyllis Lepperd. I am the Chief of the Reproductive Sciences Branch. I want to thank Dick. Dick has been a member of the Reproductive Sciences Branch longer than any of us and has had a lot of experience and was the person who really was pioneering in the use of the cooperative agreement in reproductive sciences.

One of the nice things about our branch, I believe, is that everyone in the branch has had experience in academics or in public health and has had actual experience either at the bench or in clinical epidemiology.

That, I think, is a real strength for all of us because we know what it is like to be out there.

It gives me great pleasure now to introduce the moderator of the panel for this morning, and that is Dr. Linda Giudice. She is a Professor of Gynecology and Obstetrics at Stanford University where she is the Director of the Reproductive Endocrinology and Infertility Program. She is well known to most of you, I think. She is very active on NIH study sections. She is also the coordinator of the Reproductive Medicine Network and has led us forward very well.

Linda?

Science and ART

DR. GIUDICE: Thank you, Phyllis, and good morning everyone. I would like to thank Phyllis, first, for inviting me here to participate as a moderator. I think it is really important to acknowledge the unprecedented gathering and interaction of the agencies that we saw this morning in putting together this program which acknowledges the importance of ART which, from a clinical perspective, is well known for its abilities for

fertility therapy although some of the other applications often get more press.

Today and tomorrow, this group has brought together clinicians, researchers, patient advocates, policy makers, political scientists and ethicists. I think this is really an unprecedented meeting and will really result in some important conclusions.

This morning's first session is entitled Science and ART. Our first speaker, Dr. Gerry Schatten, is from the Pittsburgh Development Center. He is the Director of the center. He is also the Deputy Director of the McGee Women's Research Institute, Professor and Vice Chair of OB-GYN and Reproductive Sciences, and also of Cell Biology and Physiology at the University of Pittsburgh.

He will begin the session by addressing What Do We Need to Know about ART; Preclinical Research and Clinical ART Practice.

Gerry?

What Do We Need to Know Now about ART:

Preclinical Research and Clinical ART Practice

DR. SCHATTEN: Thank you, Linda, for that kind introduction. I want to also echo the words that the

previous speakers have said regarding the historic nature of this meeting here today. While I am setting up my computer, I want us to remember that we are here because of the successes of the past quarter century.

We are here because there are a million souls on earth who wouldn't have been here were it not for the extraordinary work that the clinical community, the basic-researchers and our colleagues here in the Beltway have sponsored.

I think we have done an extraordinary job. I think we need to look forward to the next quarter century and, as in the past one, exceed all expectations and do an even better job. I must say, I am touched by the spirit of the statements that have been made because it would be very easy to see how folks in the room might argue that people on one side of the Beltway have one view and those on the other side of the Beltway might have a disparate view.

Were that to be the case, everyone would lose and, perhaps most importantly, the couples who are seeking desperately to have children and to have healthy children would lose. I certainly want to underscore that fact that we are all in the same boat. I think there are some

straightforward steps that we can take to continue to move us forward, but we need to be strategic, we need to be sensitive and we need to be thoughtful on that.

So I have titled the talk, What Do we Need to Know Now about ART. Many of you know I happen to be a monkey's uncle. My nieces hate this, but I do want to speak a lot about preclinical research and clinical ART practice.

[Slide.]

Dr. Tasca spoke about, in a sense, where we are now. I want to just highlight that a little bit more. We know some aspects of spermatogenesis. We know some aspects of oogenesis, but our knowledge of the way the gametes are constructed is still in its infancy. I commend our friends at the NIH for their focus on female health and what that does for egg quality.

You know, a quarter century ago, we have the first IVF babies in which sperm, through some mysterious mechanism but one that mirrors the in vivo situation were selected and that single sperm would outcompete its brethren and get into the egg.

Just ten years ago, an extraordinary accidental discovery was made in humans. You all know that as intracytoplasmic sperm injection where you could take a single sperm and inject it into an egg and have an extraordinarily high rate of fertilization, of embryogenesis, of pregnancies, of babies. The oldest child now is ten years old. I don't know whether it is 25,000 children, either having been born or gestating, or more now.

This I need to remind you was discovered in humans and had to be discovered in humans. Had those clinicians read the literature better, they would have recognized that it would never have worked. It doesn't work in many species and it is only now, after the human successes in ICSI, that some of us have attempted ICSI in monkeys where it works just as well as in humans.

From that work, others have now performed and failed and revised and improved techniques so that ICSI does work now in mice and in cattle. This is an example of translational research but it is from the bedside to the bench.

The basic-research community needs to acknowledge what privately funded human ART studies have done for basic research. Were it not for the privately funded ART experiments on patients paying to have it done on themselves, we would never have had the surplus embryos from which embryonic stem cells have been derived.

This isn't an example where the NIH chose to abdicate its role. This is an example where the NIH was forced to abdicate its role. Again, I don't want to belabor the embryonic stem-cell implications, except to say that, within the broad context of human embryology and human fertilization, all aspects of medicine come into play in part because of the promise of embryonic stem cells.

Dick already spoke about cytoplasmic transfer. We will talk a little bit more about that. There are issues that we don't fully yet appreciate about--ELSI, here, for the cognoscenti, is elongated spermatid injection. It is not ethical-legal stuff yet, but it will get there.

There are questions about in vitro culture, genomic imprinting, DNA damage repair, issues about mitochondrial transfer. We also know now, because of

preimplantation genetic diagnoses, or what is also referred to as aneuploidy screening, that infertility therapies are now being offered to couples of advanced age.

I hate to say that advanced age for women is just so young. Here I am a geriatric old guy and a woman in her 30's can be called old. It is incredible. As a result of that, we know more and more about what goes on in terms of removing a blastomere. But there is a lot of basic research in this area that still needs to occur.

I put up here, in additional to the implications of embryonic stem cells, questions about cryopreservation. Cryopreservation is something that we have taken for granted but we really don't know yet enough about it. Yet, you can read on the airline magazines that people can freeze ovarian tissues. I don't question that the freezing of ovarian tissue is, indeed, a reality. I think more work needs to be done, though, on the thawing.

The future here is promising, but we are in an area that captures a lot of attention and there is a danger to have the hype coming before the hypotheses.

[Slide.]

I want to outline my talk in this way. I put, in this case, the ELSI issue on the top because I find that, so often, if we end up putting the ELSI issues on the bottom, you ending up grappling with the ethical issues after you have completed the work when, in fact, I think, for all of our sakes, we are better off in having the ethical, legal and social conversations up front continuously and after the fact.

I think so many questions we can reach consensus on through that whole process. I think we are in a magical opportunity right now because, for the first time in my lifetime, I think the American public has seriously grappled with what the importance of a human embryo is and seriously grappled with the issue of whether a human embryo in a plastic dish is identical to a human embryo that is within the womb of a prospective mom.

I so hope that we all can build on the current momentum and continue knowledgeable discussions so that the public's attention can be focused on the serious issues and not the science-fictional aspects of cloning and things like that.

I will speak a little about ART, but I think our main challenge today really deals with safety and efficacy and there are some retrospective clinical-outcome studies, many of them very good. Where I think we need to look forward in this next quarter century is on prospective studies.

I don't know if this is the right acronym, but you know how there are SWAT teams for, I don't know if I can say the word "anthrax" in here, but SWAT teams for whatever. In a sense, I think we need to do targeted preemptive research. Maybe SW-ART is the wrong word, but basically a SWAT team for ART so that, when an innovative therapy, say cytoplasmic transfer, is proposed, it isn't a half decade or more before basic researchers digest the information that has been published, submit their grants, hear from Dick that they are going to be renegotiated out of business and appeal to Phyllis and maybe rattle a few extra nickels out and then finally start experiments by the time those offspring might already be in grade school.

We can expect that the field is moving rapidly already and it is only going to accelerate. I think, as

community, we can be smarter and invest our resources, intellectual, physical, financial, better.

There is an irony that one aspect of the Center for Population Research focuses on things up to implantation and then things move over into postimplantation worlds which, I think, falls into the Mother's and Children's Branch. Because ART is unique in having generational consequences--there is no other aspect of medicine that has the same kind of generational consequences--maybe we need to be looking at our lives not as buckets where you think about the kidney or buckets where you think about the heart, but maybe we need to think in terms of the full life history and somehow merge the events of gametogenesis and preimplantation development with fetal development and with neonatal pediatric and adult outcomes.

As we heard earlier, transplantation medicine is right in the middle of where we all sit. Whether we want to call this in the context of stem cell biology or cloning or transgenics or germline gene transfer after gene therapy, these areas are intimately entwined with appropriation right now. Certainly, there are issues about

infectious diseases. There are issues about what can or should be done with embryonic stem cells.

I was told not to use four-letter words, but I will say cloning. There is great interest in doing therapeutic cloning. Unfortunately, there is so much sensationalism from charlatans on reproductive cloning, and I will touch on that, if you wonder what I think.

[Slide.]

Many of us are here in part because of cytoplasmic transfer. Cytoplasmic transfer was and actually remains an interesting innovative therapy for couples who have experienced repeated IVF failures. The notion is that a donated egg might have cytoplasm that is superior to what I call the challenged egg. I was trying to be politically correct.

You introduce some of this ostensibly younger highly-charged cytoplasm into the egg when you inject the sperm and you end up with a healthier fertilized egg and embryo and some offspring. It is unclear whether the technique really has the efficacy that had been proposed. There are concerns about mitochondrial DNA inheritance, and

we can talk about that more later, but I think, in a sense, cytoplasmic transfer highlights where we have failed.

It highlights where we have failed because, in our country, we have two disparate worlds. These worlds are represented in this audience. We have a world of privately funded clinics who have helped the world have a million babies. For those of you that trained in surgical specialties, you know that there are risks in every medical procedure and the acceptability of risk from those of you coming from clinical subspecialties are different than those of us who learned about molecular biology in worms or in yeast or in flies.

Our sense of risk and our sense of what is compelling conclusive discovery is different than the evidence that people in the surgical world will bring to bear. So, in many ways, I think we may be communicating using the same words but the words aren't hitting because they have very different meanings to us.

If you ask a basic researcher what is the evidence, say, on cytoplasmic transfer, your answer might be, well, we need $50 million and thirty-five years and we will get back to you. Needless to say, if you are bringing

a baby that is convulsing to an emergency room, god forbid you have a Ph.D. debating how you know whether or not the baby is really there.

So I think it is important to put on the table that each of us bring in different issues and different criteria for what we mean by the very same word.

[Slide.]

So I want to speak a little bit about the problems are garnering relevant evidence. We are here to talk about evidence-based ART. There is an enormous problem with this because the evidence isn't available yet. One of the reasons is that fundamentally all different programs practice in different ways.

So even when we see an extraordinary paper coming out, say in the British Medical Journal or in the new England Journal of Medicine, you will see many of us will debate the merits of the paper simply because of the underlying assumptions that occur in each different practice.

There are variations in data collection and others can speak about how complicated data is reported and these complicate the interpretations. Also, ART is moving

so swiftly that we really don't have time to do the careful retrospective analyses that you really want for solid evidence.

In fact, every study that comes out frequently is criticized that, oh my god, yeah; that is the way they did it in '98 but, come on, it is 2002 and we are no longer doing that way, so it really doesn't apply anymore.

The procedures are technically demanding and individuals have different expertise and instruments. There are steep learning curves. So, again, it is hard to compare apples to apples. Again, there are innovations that are quickly abandoned and replaced with newer approaches. Furthermore, as infertile couples cannot wait that thirty-five years to see if something really is successful, there is a swift time frame here.

[Slide.]

So, while I think it is commendable and appropriate to be talking about evidence-based ART, I think it is challenging for a number of reasons and including not only garnering the evidence, but disseminating the evidence. ART practices in our country, unlike those in other countries, are entrepreneurial. There are issues of

scientific and medical competition. There are financial competitions.

Without focussing maybe only on the New York area, we can say, does Macy's tell Gimbels, or does New Jersey tell New York, or whatever. Those competitions, which some of us are not involved in, are real to those folks who are in those competitions and are being told, look, you are not close to zero; you are not at the bottom line.

There are also technological issues and I want to commend the NICHD for helping fund some advanced training courses like Frontiers in Reproduction at Woods Hole. There is also proprietary information. Some of that proprietary information may fall in the category of intellectual property or patents, technical know how, and it doesn't matter whether you are in the ivory towers of Nirvana, as I am, in an academic institution, or a private program or in a company.

Each different unit has its own way of handling, or mishandling, intellectual property and that also forestalls the swift translation of information. So even

if we could garner the information, I am not even sure we could disseminate it as swiftly as we want.

So I need to tell you about monkeys.

[Slide.]

We got into this business about a decade ago, I guess, through Dick Tasca and Phyllis Lepperd and Florence Hazeltine and Duane Alexander's help, basically extrapolating IVF methods or ART methods that were successful in mice to monkey and then, from the clinical world back to monkeys, working with testicular sperm. You can see Tess and Ticker.

We have a whole host of primates that we have made through various ART programs. The purpose of this initially was because we were being criticized that using nonfederally funded research facilities and nonfederally sponsored studies with informed consent and Vatican approval, we were looking at inseminated human eggs that had failed to fertilize and were clinically discarded and we were discovering some aspects of human reproduction that differed from other animals.

But cranky members of the audience--I don't know; maybe like Lou DePaolo, would say, "But look, Gerry; you

are looking at failed material from infertile couples. These couples showed up because they were infertile, so the material might have been compromised and, plus, they failed. The material you got was the leftovers, so it was failed."

So, certainly, there were reasons to doubt the accuracy of the studies because you were looking at discarded material from infertile patients. So, basically, we set up IVF and ART in primates, initially in Wisconsin with Barry Bavister's help and John Hearn.

[Slide.]

The purpose of this was to now use perfectly primed breeding rhesus monkeys to ask questions about what happens during ART. I am going to show you an animation that Laura Hewitson has put together for your purposes here about ICSI.

[Animation.]

The way ICSI works is you basically use a very fine needle. You aspirate a single sperm into the fine needle. In the current administration, we no longer us the word "suck." A fine needle, a polished needle, is used to

hold the human or monkey oocyte. The needle is injected into the egg by breaching the plasma membrane.

We don't understand how that ICSI sperm really activates the human or primate egg, but it does activate the--the meiotic spindle completes its work. The second polar-body nucleus is out there. The egg nucleus migrates down a complicated microtubular-based structure to reach the sperm nucleus.

The are some choreographical differences between ICSI and IVF. One of them I will refer to as a condom, but I will tell you about that in a second. The sperm in humans and in primates brings in the centrioles which forms a three-way system of microtubules that brings the egg nucleus to the sperm nucleus. It duplicates and then first division occurs.

[Slide.]

In humans and nonhuman primates, not in mice, the centrioles in the fertilized egg come from the father. So, for those of you that remember your mother on Mother's Day for the mitochondrial DNA contribution--remember all that?--please, in the spirit of gender equity, on Father's Day, centrioles.

Here is a discarded human oocyte in which you can see two spots of gammatubulin, which is a rare tubulin marker for the centrioles, that comes in with the sperm tail. There are certain forms of infertility where the man's sperm has centriole defects. The man's sperm will enter the egg but it will not form the structures that bring sperm and egg nuclei together. So there are novel forms of male infertility that are only resolved after the sperm is in the egg.

[Slide.]

Later, and this is also a discarded egg--this is the sperm nucleus with this freeway system growing here. The egg nucleus here, second polar body. I think it is important to realize that human eggs are not dissimilar from eggs of other mammals or eggs from Drosophila or C. elegans and that, from a basic molecular and cell-biological foundation, they all look so similar and they use the same biology.

[Slide.]

ICSI differs from IVF in some small ways. Whether those small ways are meaningful or not is a challenge for us in the future. One thing is when the

sperm comes to the egg, in a sense, the sperm has a necktie around its apical region. This necktie is shown here in an ICSI monkey.

The necktie is made of membrane fusion proteins which, when the sperm fuses its membrane with the egg, it is as a fusion-protein molecule. These necktie molecules enter the egg's plasma membrane and diffuse into the egg's membrane.

In contrast, during ICSI, this membrane structure, this necktie, remains around the sperm head and the decondensation of the male pronucleus is retarded.

[Slide.]

As a result of that, you can see already, as the *spermaster is growing, the apical region of the sperm head has not swollen at the same velocity as the basal region. Ironically, the X or the Y chromosome, the sex-determining chromosomes, are nonrandomly positioned in this apical region and there are questions of whether some sex chromosome anomalies seen in ICSI pregnancies could be the result of the ICSI procedure, itself.

[Slide.]

There is also a condomlike structure on the apex of the head. You can see that here. This condomlike structure is lost typically just after the sperm enters the egg cytoplasm. The BBC, by the way, wanted me to refer to it as a "hard hat." Those British are so proper.

That condom stays on the male pronucleus longer after ICSI and the initiation of first DNA replication is delayed after ICSI as contrasted with IVF. Does this matter in the big scheme of things? There probably is a small influence. Whether this is the same influence of the gravitational force of Jupiter when you were born or larger, I don't know.

Actually, we should know some of these answers and we should know them not only for ourselves but for the community and for the American taxpayer.

[Slide.]

Here is another image showing that same event where this condom is on a sperm nucleus as it is decondensing in a rhesus egg and the choreography of decondensation differs slightly.

[Slide.]

Later, the two pronuclei come together and you form the first mitotic spindle. This is also in a discarded human oocyte.

[Slide.]

So there are some choreographic differences between the ICSI and IVF and the jury is still out about whether these are clinically meaningful. I think these are among the bits of evidence that we need to garner through a cooperation among clinicians and preclinicians.

[Slide.]

There is another question and that is sperm selection and the mode of sperm entry. We don't really understand why the one sperm that enters the egg is selected by the oocyte. Has it to do with motility, penetration through the zone, the acrosome reaction, binding to the egg membrane? We just don't know.

Nowadays, we look down the microscope and someone says, "Ooh; that's a cute one," and selects a sperm using, perhaps, capricious criteria. The women in the room might want to look around. Maybe everybody would agree that Professor Biggers is the best stud to mate with and he would be the one selected to father your children.

Some of us might disagree. Right now, I think we are in an awkward position because we don't have those criteria to determine which of those sperm should be the one selected to father the next generation. I can see that the women agree that Professor Biggers should be the one.

[Slide.]

There is another issue and that is that foreign DNA in an experimental circumstance can bind to the outside of sperm. So, here, looking at the rhesus monkey, you can see rhodamine conjugated, DNA bound to the outside of a rhesus sperm.

[Slide.]

You can see it here with the ICSI needle. You can see it here with the DNA bound to that sperm being injected into the oocyte.

[Slide.]

We put in a plasma that expresses the green fluorescent protein and, already by the four-cell stage, you can see transient expression.

[Slide.]

By the blastocyst stage, you can see that there is expression of the green fluorescent protein in the

inner-cell mass cells as well as in the troph ectoderm, trophoblasts.

By the way, there is a polarity to the human embryo as well as the mouse embryo and the polarity is being discovered by brilliant investigations primarily from colleagues in the U.K., Richard Gardner, Roger Peterson and Magdelena Zernicka-Goetz--I always have a problem pronouncing her name. These have implications about where you inject the sperm because it is likely that the site of sperm injection or sperm entry determine one of our embryonic axes.

You know we have a top and a bottom and a belly and a back and a left and a right. It may well be that the position of either sperm entry or sperm injection determines the left-right axis. The implications of this for either ICSI injection or even preimplantation genetic diagnosis when you are doing embryo biopsy needs further investigations.

[Slide.]

This is George. Let me leave that.

[Slide.]

We have heard about gene therapy. In a sense, there is an area I believe should also be called into play and that is whether there is a possibility of germline gene transfer after gene therapy. If you think about ART, in some ways, germline gene therapy mirrors some of these events. I don't want to belabor all of these. Some of these may be in the handout, but I did update the handouts.

There are both intentional as well as unintentional questions about germline gene transfer during ART. In a sense, there is an intentional manipulation of the genome during PGD but that may be a different example. I do think that questions about gamete alterations after gene therapy is an area that has been understudied and requires significant attention.

[Slide.]

Last year, we were involved in publishing some work on Andi, a monkey that has inserted DNA and we use the term, the reverse acronym, inserted DNA to come up with Andi's name. A fetus that was stillborn had green fluorescent fingernails and toenails and hair and fluorescental placental tissues. While we were encouraged to go for a small business grant with Mabelline, we do

think that what we are doing in making transgenic primate models to accelerate molecular medicine from the mouse world to patients does, indeed, have implications because it is a step towards proof or principle for whether you want to call a germline gene therapy or genetic enhancement.

I don't want to minimize the ethical consequences here. Furthermore, there are compelling reasons to do transgenic experiments on human embryonic stem cells. But the discoveries that we make on how to modify genetically the human embryonic stem cells will be the foundation on which others may use the same information to genetically modify human embryos or human gametes.

So I don't want to minimize that.

[Animation.]

Again, let me give you another brief animation. The fields of gene therapy and genetic enhancement are going to come together faster than any of us want. The way that we made Andi and his brethren was to use a gutless retroviral vector. This is something familiar to those of you in the gene-therapy world. It was introduced between the egg membrane and the outside zona pellucida.

Of course, retroviruses come in as single-stranded RNA which are reverse transcribed into single-stranded DNA. By the way, if you see Spiderman--I will tell you about that later--it becomes double-stranded DNA. You know, Spiderman isn't a radioactive accident anymore. He is done by some idiot scientist using gene therapy. You can see aspects of this animation in the movie.

But, anyway, the foreign DNA enters into the blueprint of the egg. Because retroviruses only insert when there are chromosomes at M-phase decondensing, it is a perfect time for the foreign DNA to enter into the maternal lineage because the female meiotic spindle is arrested at second meiotic metaphase. The DNA enters there. The sperm comes in here and I wish I had had the sound track for Jaws. The embryos develop as they normally would have and then the rate of implantation is the same.

[Slide.]

An area that I think we can move into in the future which is extraordinary is noninvasive in utero imaging. This is work done in cooperation with Carnegie Mellon University where we can perform high-resolution, three-dimensional, imaging of fetal development in utero.

So here is one mouse fetus in utero, another one. You can trigger the images on the basis of collecting it in utero.

I think there is a huge gap from implantation to birth that can now be filled in.

[Slide.]

There are aspects of embryo dissociation-reaggregation. You can make artificial twins, embryonic stem cells.

[Slide.]

I do want to spend just a minute on two other points. One of them is outcome studies. We have been following fifteen animals--I'm sorry. We have been following newborn monkeys where we have fifteen controls, five split embryos, four ICSIs and four IVFs. I just want to mention that, as we follow these monkeys for psychosocial behavioral motor, other issues.

In all cases, the ART monkeys seem to display mild aspects of hyperactivity; that is, there are greater behavioral changes and we can measure that through--this is the IVF controls which are not highly significant but, in terms of split embryos, in terms of ICSI embryos, there is greater distractibility in these primates. Again, you can

see that these animals change their behaviors far faster than the controls.

[Slide.]

These animals, and animals that Maurizi Bartolome* and Richard Schultz are making at Penn, I think, in some ways, are the behavioral canaries in the mine.

Sorry; clones. I will leave that alone.

[Slide.]

I think there are ways in which this community could set up targeted research which would establish a consortium among clinical practitioners and researchers like the Culture Club where there would be a very large cooperative agreement involving a number of visionaries and practitioners to identify what the knowledge gaps are, prioritize them, ascertain the cost and then enable the conclusive studies.

The American public deserves better answers and we don't yet have them. I think our challenge for the next twenty-five years is to get those answers as swiftly as possible.

Thank you for your attention.

DR. GIUDICE: Thank you very much, Gerry, for a thought-provoking and, as always, entertaining presentation.

We will have time for discussion later so I would like to move on to our next speaker who is Dorrie Lamb who is the Director of the Laboratory for Male Reproductive Research and Testing and Associate Professor in the Departments of Urology and Cell Biology at Baylor College of Medicine.

The title of her talk is Types of Abnormalities in ART: Consent and Follow-up Issues.

Types of Abnormalities in ART:

Consent/Follow-up Issues

DR. LAMB: Thank you very much.

[Slide.]

I actually have a very difficult topic to talk to you about because there are some major issues associated with trying to even do research on ART. This is because we know that the whole process of reproduction is extremely complex. It is not just the events of fertilization that we need to be considering but all of the steps involved in gamete production, transit of the gametes as well as

fertilization, implantation and early embryonic development are necessary for us to consider in any type of study of ART.

[Slide.]

Importantly, we have to consider the fertility potential of the couple as a couple because this fertility potential can vary depending on who the partners are of this couple. So a couple may be infertile together, but with other partners may have a different fertility potential.

So, as you can imagine, this increases exponentially the complexity that we have to evaluate in terms of our assessment of the safety and efficacy of ICSI and other reproductive techniques. We have to, therefore, consider the male and the female factors, the cause of the pregnancy, as well as the offspring.

So, clearly, the challenges associated with this type of study are very significant.

[Slide.]

Clearly, there is a need for studies on the safety of the ICSI procedure which is the work that we are involved in with regard not only to major malformations and

chromosomal abnormalities but to look, overall, at the effects on the offspring.

The study that we are funded by NICHD to perform focuses on the molecular and clinical analysis of the male factor couple and their ICSI-conceived offspring.

[Slide.]

So, to do this, we assess the female factor, the male factor, the pregnancy and the offspring.

[Slide.]

To begin with, we have very definite informed-consent issues. One problem, I think, with trying to get informed consent to do this type of study is that the science and biomedical background of many patients is sufficient. We are all well aware of the deficiencies of science education in our schools. Although many ART couples have really learned a lot about their infertility, they still don't have the type of understanding to understand the basic research proposed and the goal of the research.

I don't think that patients really understand how little we know about the causes of their infertility. To give you an idea of the depth of this, just our normal ICSI

consent form that is given to our patients, not for research but just to have the procedure done, one of our physicians said, "Well, we don't want to tell them all of this, what we don't know, do we? They might not want to do this."

So you can see that there is a real lack of understanding, I think, in terms of our knowledge of ICSI and the causes of the infertility that require these couples to undergo ICSI.

Also, the couples who are being asked to participate in these studies of ART desperately want a child and so they may sign anything associated with this goal. This is also, I think, a very negative aspect in terms of trying to get informed consent.

[Slide.]

For many couples, the male partner may rarely be present for the counseling and the consent. Sometimes, the men say, well, I can't take off from work. I need to do this with a notarized signature on a consent form. In our experience, the male is more likely to sign a consent for research if he is present with his partner than if he is present alone.

I think that is because women, in general, seem to be more motivated in terms of healthcare issues.

[Slide.]

Then we have big problems that we face with informed-consent issues due to the guy factor. I hope the men in the audience will excuse me for this and maybe this is typical of Texas men, but the men in our experience frequently refuse to participate in research studies. I was astounded, actually, at the numbers who choose not to participate.

Some of the comments we here is, "What's in it for me?" "Will I be paid?" No. "I faced this challenge and my kid can use ICSI in the future to beat this problem." "If it doesn't help me, why should I do it?" Then a big concern is, "Will you clone me?" because we are looking at genetic aspects of male infertility.

Interestingly, we also find that, depending on who asks these patients to sign the consent, we actually have a very different rate of agreement. So when my male urologist fellows approach these patients for consent for research, usually they refuse.

I don't understand this, but I now have a young lady physician who has been doing an excellent job and, for some reason, the men are much more willing to sign the consents for her. Again, I don't know what it is but there are major differences just based on who asked them to sign the consent.

[Slide.]

What they are consenting is to allow us to have access to huge amounts of information which I have listed for you here. You can see that we are asking for lots of types of information of the male patient, the female patient, analysis of pregnancy and fetal studies as well as postnatal studies.

Each one of these categories that I have listed for you isn't a single input piece of information. So, for example, for semen analysis, we are getting count, motility, morphology, DNA damage, immunologic infertility kinds of issues. So each one of these categories has a huge amount of information in the database so you can see that there is enormous data-gathering information that requires a huge amount of effort by our group.

[Slide.]

So there are a lot of challenges associated with trying to get this information. One of the main ones is that, at least in our experience, infertility patients go to large IVF clinics in major metropolitan cities to achieve a pregnancy by ICSI.

What happens is that then they return to their local OB-GYN for prenatal care and we have a very difficult time to follow them. I think this is common to many ART centers. In our experience, we have a lot of patients who come to my colleague, Larry Lifschultz, from all over the world. So we have patients from Mexico, Saudi Arabia and the Middle East in addition to patients coming from all over the United States.

So you can imagine there are very significant challenges with trying to follow these patients.

[Slide.]

There are also multiple physician surgeons in ART centers that are used by the patients who come to us for the male-factor treatment. For some of these ART centers, research may be their priority. So we, again, have some difficulty trying to collect the data.

Female classification on the female side may not be consistent because we have all of these different ART centers that we work with and importantly, as Dr. Schatten mentioned, not only may laboratory results vary markedly between clinical labs but between, different embryology labs, there are very different practices that go on.

For example, one of our embryology labs does a blastocyst transfer. Others transfer earlier on. And they all have very different fertilization and pregnancy rates and very different patients that they even accept into the program. While we take into account all of this confounding information, you can see that it, again, increases the complexity of what we are trying to look at which is the safety and efficacy of ICSI.

[Slide.]

We also have experienced great difficulty to follow fetal loss. Again, because we have out-of-town patients, it is difficult for us to get materials from these aborted fetuses for further analysis. For the couple, they have tried too hard to achieve a pregnancy that this is an emotionally devastating event for them and so they choose not to cooperate or they may simply forget

about informing the research coordinator in a timely way for us to be able to follow up these patients.

So, again, we have great difficulty trying to follow the pregnancy.

[Slide.]

The follow up of the infants is also challenging although we do have a phone system where we talk with the couples, we talk with the pediatricians. We have forms for the pediatricians but we also have examinations which are required of the infant. But, again, we have out-of-town couples who must travel to the site for the analysis.

We have working parents who can't afford to take off from work for additional medical examination of their children even though there is no cost to them. They may not want to find that there is a problem with their children and there may be a general lack of compliance in spite of the fact that the patients did sign earlier on before they became pregnant.

[Slide.]

The difficulties that are most significant, I think, are the diagnosis of idiopathic or unknown etiology because it is very difficult, then, for us to even know

what to follow up for patients in terms of looking at the offspring for any types of abnormalities.

Many of these cases represent a genetic black box with the potential for multiple gene defects resulting in a similar phenotype so there may be very subtle differences between patients, for example, with a meiotic arrest but the genetic cause may be very different.

[Slide.]

To give you an idea of the magnitude of this type of problem, I show you here the distribution of final diagnostic categories found in Larry Lifschultz's fertility clinic. We found that nearly one-quarter of all patients were diagnosed with having idiopathic infertility as shown here and, importantly, even some of these patients with varicocele could probably be included in this idiopathic infertility group, so you can see that this is a very significant number of male-factor patients.

[Slide.]

Therefore, you need to have a very expert diagnostician and consistency in patient classification looking at very subtle phenotypic differences between the patients in terms of our clinical assessment. We also,

then, have to do a very in-depth molecular assessment to try to determine the cause of these couples' infertility.

[Slide.]

Many couples, we believe, both male and female, may have genetic defects leading to their infertility. It is thought that, by bypassing biological barriers that are normally in place to block fertilization by either defective sperm or eggs, that there may be an increased rate of congenital defects but, importantly, there may also be long-term systemic consequences for the offspring that we don't even know whether or not to focus on yet.

Certainly, it is likely that at least some of these reproductive defects may be passed on to the fetuses.

[Slide.]

So the questions that remain to be answered regarding the safety of ICSI are, will men and women who are genetically programmed to produce abnormal gametes conceive children who will be infertile. As Dr. Schatten told you, the oldest ICSI baby is only about ten so it is going to be another ten to fifteen years before we can even begin to assess this question.

Perhaps, more importantly, do genes that cause poor fertility cause other systemic problems later in life. Obviously, we don't know the answer to this but it is an issue of concern and I am going to give you some of the evidence for that concern.

[Slide.]

We know that there are many genetic causes of male and female infertility. These are chromosomal defects as well as gene deletions or mutations.

[Slide.]

I would like to give you just a few examples of why we are so concerned about these genetic defects. I show you here a normal chromosomal complement, in this case for a male karyotype. Up in the upper left-hand corner, you can see a different way of looking at the chromosomes with a spectral karyotype. We know that normal men and women have twenty-two sets of autosomes and one set of sex chromosomes, so the females are XX and the males are XY. You can see here the X and the Y chromosomes.

To give you an example of what type of information this analysis is giving us, this would be like looking at the outside of an entire set of the Encyclopedia

Britannica and saying, yes; all of the volumes are present so, therefore, we think the information is okay. So you can imagine this is a very gross assessment of the genetic state of the individual.

[Slide.]

However, even on this very gross level, we know that chromosomal abnormalities are increased in infertile men as compared to fertile men.

[Slide.]

Most of these sex abnormalities are in the sex chromosome.

[Slide.]

Indeed, Klinefelter syndrome, which is having extra X chromosomes, accounts for a very significant proportion of all of azoospermia. However, if you don't screen the individuals for these chromosomal abnormalities, you will never know that they are present and, indeed, many males and many IVF centers are never screened before ART procedures are undertaken.

[Slide.]

Just so you don't think that the men are the only ones with these chromosomal abnormalities, this is data

taken from a fairly recent paper by Gekas where they show that looking at ICSI couples that not only do the men have a specific incidence of chromosomal abnormalities, the women do as well.

[Slide.]

If we look at these abnormalities by type, we can see that, although there are some gender-specific differences in the types of chromosomal abnormalities that are present, the take-home message is that, again, this is a significant problem that we need to be worried about in terms of our patients but frequently are never diagnosed.

[Slide.]

In addition, we also know that Y chromosome microdeletions, many of them too small to be seen at this karyotypic level, can cause some cases of male infertility and, although the percentages vary between about 8 to 14 percent of azoospermic men, it is important to keep in mind that, again, this is another genetic cause of male infertility.

[Slide.]

What are the clinical implications of this? The implications are that these chromosomal defects will be

transmitted to the offspring. Indeed, ICSI offspring have a higher incidence of sex chromosome abnormalities, but, many times, the patients are never evaluated for chromosomal defects to begin with. They simply undergo the ART procedure.

[Slide.]

Just to give you a further look at the concerns for these genetic causes of male and female infertility, this slide happens to be taken from a paper that Marty Matzuk and I have in press in Nature Medicine coming out next month that reviews the genetic causes of infertility. This slide happens to show you male infertility.

As you can see over here are some of the chromosomal defects that we discussed, but you can see that, even today, we do recognize that there are a number of different causes of male infertility that have a genetic basis for sex determination and development, various endocrinopathies, problems with sperm production and function. There are actually more than are shown in this slide that have to do with obstructive azoospermia and other types of sperm defects.

The point is that there are a significant number of genetic causes already identified. For example, myotonic dystrophy, which you see up here under sperm production and function, recent studies in my laboratory have shown a very high percentage of the nonobstructive azoospermic patients to have undiagnosed myotonic dystrophy in a milder form.

So, again, there are concerns because patients are not evaluated for some of the different genetic problems.

[Slide.]

To give you an idea of the magnitude of the concern, because we, in reality, know very little about the genetic causes of infertility in the human, I have summarized for you here a number of different genes which are known to be required for fertility in the mouse. These were all studies using targeted gene deletion. For all of the genes that I will showing you, the mice were infertile. They were not subfertile and there were no combinations of different crosses done to achieve the infertility.

[Slide.]

So here I have listed for you the various genes in the mouse which are involved in--that affect both sexes. Again, genes involved in sex determination and development, whether or not you get a gonad, whether or not you get a testis or an ovary and so on.

Endocrinopathies; genes that affect meiosis in both sexes. Hypogonadal animals with small gonads and other reproductive defects. So you can see already there are quite a few genes.

[Slide.]

If we look specifically at male infertility genes affecting only spermatogenesis, in the center you can see the cells that are affected, the various processes which are required for spermatogenesis which include mitosis and meiosis in the testis as well as differentiation of the cells into, finally, mature spermatozoa.

What I have listed on either side are the genes which, again, when knocked out in the mouse, cause male infertility due to a spermatogenic defect. I would like to emphasize that there is a large number of these genes which are listed.

Every week, as Marty and I tried to prepare this paper, we kept having more and more genes to add. So this is a rapidly advancing field but the take-home message is that that there are many of these genes which are required just for spermatogenesis, not any of the subsequent processes required for transport of the sperm or fertilization and so on.

[Slide.]

Again, for the female, there are many genes which were knocked out in the mouse, caused sterility. I have shown you here a listing of the genes that are known so far that are required for follicular genesis and ovulation as well as fertilization, preimplantation development and implantation.

So, again, the concern is that there are many genes which we do not have the power, right now, to evaluate in the human and the translational work has not been done.

[Slide.]

Why are we so concerned about these? If the animals are simply infertile, then, perhaps, we can simply

use an assisted reproductive technique to overcome their sterility.

We know that there are systemic consequences of some of these gene mutations. I show you an example here of a group of proteins that we are working on in my laboratory which are the DNA mismatch repair proteins. We know, again, in these mouse models that when these genes are knocked out, we get abnormal spermatogenesis with meiotic arrest.

In cancers, we know that it is a cause of microsatellite instability when these genes are knocked out and, indeed, patients who have a mutation of these various genes have associated with this mutation problems with early cancer developments such as hereditary nonpolyposis colorectal cancer, ataxia telegestasia, the development of lymphomas and sarcomas at very early age. This is in addition to the fact that they have male and female infertility.

[Slide.]

We have a great concern because there are many genetic causes of male and female infertility. There are laboratory tests available today to evaluate and diagnose

just very few of these defects. In fact, many of these methods are not even used on patients today. There are many patients who undergo assisted reproductive technologies who have not even had a karyotype analysis.

[Slide.]

So, really, one of our main deficiencies in terms of following up the infertility couple and trying to assess the safety of an ART procedure comes from our deficient knowledge of the sort of basic mechanisms which are required for reproduction. We don't have a knowledge of the genetic basis of infertility in humans. We don't understand the molecular basis for gamete production, transit of the gametes in the genital tract, fertilization, implantation and pregnancy.

Therefore, it is really impossible for us to understand the causes of the infertility. As such, we don't even know exactly what we should be looking at to follow up the safety of the procedure for the offspring because, certainly, we can look to see whether a baby has ten fingers and ten toes and a medical geneticist can pick out some of these subtle dysmorphias that a pediatrician would miss but, again, there may be systemic problems later

in life that we don't even know to look for yet because we don't understand the basis of the infertility.

[Slide.]

So, clearly, the follow up of ICSI couples and their offspring presents a series of challenges to the basic-science or transnational-science laboratory. I think that it is very clear that there are significant deficits in our understanding of the basic mechanisms of infertility.

[Slide.]

Clearly, patients need to be counseled that, although studies to date on the safety of ICSI are reassuring, there are no guarantees of a perfect baby even with advanced genetic testing. Certainly, they should be encouraged to participate in all of these research programs to clearly define the safety and efficacy of these ART procedures.

[Slide.]

I would like to simply thank the NICHD for their support of our program project on the genetic basis of male infertility.

Thank you.

DR. GIUDICE: Thank you, Dorrie.

It is now time for a break. We will have time later for questions. So please return at 10:15. Thank you.

[Break.]

DR. GIUDICE: Our next speaker is Laura Goldsmith who is Professor of OB-GYN and Women's Health at New Jersey Medical School. She will present Endocrine Issues in ART Complications.

Laura?

Endocrine Issues in ART Complications

DR. GOLDSMITH: Thank you, Linda.

[Slide.]

I would to thank Dr. Lepperd and the organizers of this very distinguished meeting for inviting me to talk about some endocrine issues that are responsible for ART complications.

I just would like to comment that I think it is really an excellent meeting that has been put together. Multidisciplinary approaches are always productive. I who am a basic scientist with a primary appointment in the clinical department and a secondary appointment in a basic-

science department find that it is more productive for everyone when communication between basic scientists and clinicians is frequent and with the kind of detail that you need by getting people together in one room, all together, for at least a day.

So I congratulate you for organizing this meeting.

[Slide.]

It is well recognized and well established that assisted reproductive techniques result in multiple follicular development, Superovulation with gonadotropin therapy is designed to, in fact, develop multiple follicles. This, of course, results in the development and formation of multiple corpora lutea.

This is well established. We know this. This has been well documented in basic science and clinical literature. What is not well recognized, actually, is that a complication of assisted reproductive technology is, in fact, premature delivery, preterm birth.

As Dr. Lamb pointed out, this frequently is because the people who actually follow the results of the assisted reproductive technique successes are

perinatologists and obstetricians who really do not communicate with the reproductive endocrinologist sufficiently so that the RE people who make the pregnancy do not get the opportunity to follow these pregnancies.

It turns out that there is now recognized an entity of preterm birth following ART. It was previously thought that this was only due to multiple gestations. It is true that multiple gestations that result from assisted reproductive technologies do have a higher incidence of prematurity but what I am going to talk about now is actually an increase in preterm birth in singleton pregnancies.

This has not been widely appreciated and I think it is important that we discuss the potential reasons for this. We now have data which actually connects these two issues; that is, we now have data that suggests that a reason for the preterm birth is actually due to the fact that multiple corpora lutea are being formed.

What I am going to talk about today is some of our data which established that fact and give reason for the endocrinology of the corpora lutea that may be responsible for preterm birth.

[Slide.]

This became apparent to us some years ago as we were perusing the literature that there is an increased incidence of prematurity in singleton pregnancies resulting from assisted reproductive techniques. So recently we have decided to do a more thorough investigation of the literature to clarify this relationship.

Recently, my colleagues Peter McGovern and Amore Lorenz* and I have done a systematic analysis of the world literature in the English language to determine whether, in fact, this increased risk does exist throughout the world and what is actually the incidence.

So we have done a metaanalysis of these studies. We actually looked at the entire world literature from 1960 through the Year 2000. We could identify, out of some 2,000 publications that were reviewed, only 27 studies that are actually case-controlled studies; that is, there are many other publications that talk about increased risk of prematurity after ART but they were not controlled studies in which there was a control group.

We thought it important to only identify those studies which had a control group. This plot shows the

relative risk of those identified in those 27 studies. As you can see, in 25 out of those 27 studies, the relative risk is greater than 1.0. If you just look grossly, you would come up with about a twofold increase, which is what we did.

[Slide.]

But in a more systematic summary, what you see is that the median incidence in the 27 studies of preterm birth--this is in only singleton pregnancies; this does not include multiple gestations--the incidence was 11.2 in the ART group. In the control group, it was 6 percent.

So the calculations suggest that the median of these relative risks is 1.93. Again, we thought it was almost funny that when we perused the literature grossly with our eyeballs, we managed to come up with about a twofold increased incidence and when we systematically did it in a very careful, methodical way, it was about a twofold increase, 1.93.

[Slide.]

So, for us, we know that the corpus luteum is well established to make steroid hormones and was previously established to be a steroidogenic organ. It is

well established the corpus luteum makes progesterone and estradiol throughout the menstrual cycle and well into pregnancy.

What has been more recently determined is that the corpus luteum is not solely a steroidogenic organ. The corpus luteum makes a variety of other factors including peptide hormones and other factors. Peptide hormones most well studied that are made by the corpus luteum include inhibin and a peptide hormone called relaxin, growth factors such as vascular endothelial growth factor, fibroblast growth factor and epidermal growth factor and, of course prostaglandins.

You may have seen in relation to assisted reproductive techniques--frequently VEGF has been widely studied and been shown to be increased after superovulation therapy and is thought to be one of the factors responsible for hyperstimulation syndrome.

Other factors are well known to be made by the corpus luteum but less well studied in terms of clinical syndromes. Fibroblast growth factor and epidermal growth factor were actually originally identified when they were first identified as products of the corpus luteum.

What I would like to tell you about today is some studies that we have done, clinical studies and basic science studies, regarding a hormone called relaxin and how our data suggest that relaxin is one involved in the reason why there is an increased incidence of prematurity after assisted reproductive techniques in singleton pregnancies and how we have used our basic-science studies to actually develop a potential mechanism.

[Slide.]

Let me give you a little bit of introduction to the field of relaxin. As we said, relaxin is produced by the corpus luteum. It is also produced by decidua and placenta in women. The source of circulating relaxin in the maternal circulation is the corpus luteum. The decidua and the placenta synthesize relaxin. Those tissues do not contribute to the circulating levels.

Relaxin is present throughout the maternal circulation throughout the duration of pregnancy and the concentrations of circulating relaxin are determined by luteal mass; that is, in women who have multiple corpora lutea, you have significantly higher levels of relaxin.

We have done a variety of studies that well document this. I can refer you to a variety of publications.

[Slide.]

Just in summary, we can say that ovulation induction with human menopausal gonadotropins causes a mean of approximately threefold increase in circulating relaxin levels.

[Slide.]

What is important to recognize is that this increase is maintained throughout the duration of pregnancy. It was originally thought that the corpus luteum of pregnancy is only functional for the first trimester, for the first six to seven weeks. When it was thought that the corpus luteum only made progesterone, it was thought that it would only be necessary for the corpus luteum to be maintained until the placenta takes over the production of progesterone which is required for the maintenance of pregnancy.

We now know that the corpus luteum is maintained throughout the duration of pregnancy and such that if you

have elevated relaxin levels, you will have elevated relaxin levels throughout the duration of pregnancy.

What is shown here is a composite diagrammatic representation of circulating levels of relaxin in a normal group of women, spontaneous singleton pregnancies, throughout the duration of pregnancy. The upper pattern is the pattern that is seen in women, singleton pregnancies, in whom pregnancy has been achieved through IVF after superovulation induction.

Please note that this is a log scale so that these differences are substantial.

[Slide.]

So we wanted to test the hypothesis that the hyperrelaxinemia caused by ovarial stimulation during ovulation induction would result in an increased rate of premature labor or preterm delivery.

[Slide.]

In order to test this hypothesis, we performed a study in which we studied women, normal women having spontaneous singleton pregnancies and a group of women who were having pregnancies after superovulation therapy, IVF pregnancies.

These were women who were studied at the Center for Reproductive Medicine, the New Jersey Medical School OB-GYN Department Endocrine Practice site that is associated with Hackensack University Medical Center. So all women who were admitted to the program were studied and followed.

Women had serum levels of relaxin measured between six and twelve weeks of pregnancy and the outcome of their pregnancy was determined.

[Slide.]

We established the normal level of relaxin in the spontaneous singleton pregnancies at 1.18 nanogram per ml and compared that normal level to the relaxin levels in the women who achieved pregnancy via IVF. We established a cutoff for what we decided would be a very high level of relaxin, what we call hyperrelaxinemia, as three standard deviations above the mean, 3.25 nanograms per ml.

That is a very conservative estimate. It is a very, very high level. In spontaneous singleton pregnancies you rarely see levels above 1.5 nanograms per ml.

[Slide.]

We defined prematurity risk as labor requiring tocolytic therapy before 37 weeks in singletons or before 37 weeks in multiple pregnancies or in those pregnancies requiring cerclage for cervical incompetence. We defined preterm delivery in this study as delivery prior to 37 weeks in singletons or 34 weeks in multiples.

[Slide.]

This is actually an analysis of a large amount of data that was generated. I am presenting it to you for simplicity. What this allowed us to do was to separate the variables of elevated relaxin from the variable of fetal number. So what this tells you is that, with elevated relaxin levels, you have an increased risk of prematurity such that, with an odds ratio of 2.06, which translates to for every increase in relaxin, maternal circulating relaxin, concentrations of 5 nanograms per ml, you have a two-fold increased risk of prematurity.

This was independent of the risk caused by increase in fetal number which was also a significant increase in risk

[Slide.]

This study allowed us to conclude that women who have highly elevated circulating relaxin concentrations in the first trimester of pregnancy are at increased risk of prematurity and that this increased risk of prematurity was in addition and separate from the increased risk due to increased fetal number.

[Slide.]

Since the time that we did the study, there have been other studies that show an association between elevated relaxin concentrations and an increased risk of prematurity. The Petersen group in Denmark did a study using singleton pregnancies, spontaneous singleton pregnancies, and found that, in those women who had elevated relaxin, there was a significant increased risk of prematurity.

We did a study with Debbie Platek at Einstein showing that, in twins, elevated relaxin was associated with an increased incidence of prematurity in that group. Ida Vogel and the group in Denmark have recently done a very detailed and larger study which also demonstrates a significant increase in prematurity in singleton pregnancies with elevated relaxin concentrations.

We also did a study in conjunction with the Maternal Fetal Medicine Network looking at their samples taken from twins, very well characterized samples, and showed a tendency towards increased levels of relaxin in samples taken at 24 weeks of pregnancy, had a higher incidence of prematurity.

[Slide.]

The question of what is the mechanism led us to our basic science studies. We know that dilation and ripening of the human cervix involves rearrangement of cervical connective tissue and leukocyte infiltration which is a hallmark of cervical dilatation.

[Slide.]

We also know that relaxin has very pronounced effects upon uterine connective tissue in a variety of species including women.

[Slide.]

We also know that the maintenance of connective-tissue architecture requires a balance between the action of matrix metalloproteinases, which are enzymes which degrade the extracellular matrix, and they are endogenous

tissue inhibitors of metalloproteinases, TIMPs, which regulate the activity of the matrix metalloproteinases.

So we decided to use these facts to study how relaxin might be responsible for an increased incidence of prematurity by setting up a model of human lower-uterine-segment fibroblasts, which is an established model for term pregnancy cervix, and studying the effects of relaxin on these cells in relation to the role that relaxin might play in stimulating or inhibiting these factors that regulate the extracellular matrix.

[Slide.]

So the human cervix is comprised primarily of Type I collagen, actually majorly of Type I collagen, and so we looked at the factors that are responsible for the degradation of Type I collagen.

Procollagenase is converted to the active enzyme by stromelysin. Both of these are inhibited by tissue-inhibitor metalloproteinase I, TIMP I. So we used our in vitro system in the laboratory to test the effect of relaxin on levels of procollagenase, levels of stromelysin and levels of TIMP I.

[Slide.]

This first graphical representation of our data shows the results of three independent experiments, each performed in quadruplicate, actually. What you see here is the levels of procollagenase measured by Western blotting analysis and a representative Western blot is shown above. Our control levels were set at 100 and stimulation by relaxin is expressed percent of control.

A significant stimulation was effected by 1 in 10 nanograms of relaxin.

[Slide.]

Relaxin also markedly elevated levels of prostromelysin, the enzyme that converts procollagenase to active collagenase.

[Slide.]

And it caused a marked and dose-related inhibition of the endogenous inhibitor. There are many cytokines and a variety of factors that will stimulate collagenase and stromelysin, but there are few that will, in concert, inhibit the endogenous inhibitor.

[Slide.]

So what you conclude here is that relaxin actually is a positive regulator of matrix

metalloproteinases at the level of the cervix because it increases the expression of collagenase and stromelysin and also inhibits the endogenous inhibitor which would give you a net effect of an increased collagenalytic activity.

[Slide.]

We also know that a hallmark of cervical ripening is infiltration of leukocytes. These cells make a variety of other agents which would also break down connective tissue and that is one of the mechanisms of cervical ripening.

So we looked at the matrix metalloproteinase, gelatinase A, that would affect type 4 collagen digestion; that is, the type of collagen that is basement membranes, blood-vessel endothelium.

[Slide.]

We showed, in our in vitro system, that relaxin also causes a marked stimulation of progelatinase.

[Slide.]

So the net effect of this would be to increase leukocyte infiltration. So these are biochemical mechanisms which would explain and provide a mechanism, a

biochemical mechanism, by which relaxin, elevated relaxin levels, could cause an increase in prematurity.

We also are very cognizant of the fact that none of this happens by itself; that is, relaxin exists in pregnancy in a situation, in a physiological situation, in which there are other factors as well, most notably progesterone. Progesterone was also tested in the same system to determine how these factors might be working in relation to each other.

What we saw was that progesterone had either opposite effects or no effect on the same parameters that relaxin had positive effects. So this allowed us to develop a hypothesis that clearly there is as relationship between relaxin and progesterone at the level of the cervix allowing for maintenance of pregnancy when necessary and allowing for the possibility that when relaxin is elevated, that balance could be challenged.

[Slide.]

Just to show you what relaxin is doing regarding lymphocyte infiltration that can occur in vivo, I show you these data recently generated from our in vivo monkey studies. This is a different system but, in these studies,

rhesus monkeys were ovariectomized and given both estrogen and progesterone to simulate a menstrual cycle and then split into two groups. One group was given relaxin in vivo and the other group was given a vehicle control.

At the end of a 21-day period of relaxin treatment, we removed a variety of tissues, just as many tissues as we could take, and have looked at the histological and biochemical effects of relaxin in these tissues. I just want to show you these data to show you that when we looked at endometrium from these animals, we were able to clearly document that relaxin caused an increase in leukocyte infiltration into these tissues.

What you see here is data from 43 control--from tissues that we looked at, 43 different fields of endometrium and a mean level. The relaxin-treated animals showed a significantly increased incidence of leukocyte infiltration into those tissues.

[Slide.]

In summary, relaxin levels are increased in women destined to deliver prematurely. Relaxin affects the uterus promoting those types of biochemical changes which cause delivery.

[Slide.]

So what I am showing you here is a diagrammatic representation of what we now think occurs which is that during early, mid, pregnancy, relaxin action is being balanced by the action of progesterone and, in fact, other factors as well.

When relaxin levels are elevated, that overshadows this action of progesterone and other factors and, at term, although progesterone levels do not decline, it is now established that progesterone actually is declined. Recent data have shown that the number of receptors for progesterone changes and that the molecular mechanisms by which progesterone works have a decreased activity such that relaxin may be playing a role in full-term birth as well.

[Slide.]

I would like to acknowledge my colleagues, Gerson Weiss, Peter McGovern, Maury Llorens, Smitha Polejuwala, Donna Cole and Andre Wotjczuk and, of course, our support by the National Institute of Child Health and Human Development for these studies.

Thank you.

DR. GIUDICE: Thank you, Laura. Again, we will have questions at our question session at the end.

Our next speaker is Dr. John Biggers who is Professor of Cell Biology at Harvard Medical School and his lecture is entitled Culture Techniques in Animal and Human Reproductive Biology.

Culture Techniques in Animal and Human Reproductive Biology

DR. BIGGERS: Good morning.

[Slide.]

I would also like to thank the organizers for inviting me to this interesting and important meeting. I would also like to reinforce what Dick Tasca said earlier. A lot of what I am going to talk about originated in the Culture Club. I must say that the ten years that I was involved was one of the most exciting periods of my life and my career.

At the end of the abstract, I give a few references. They are all recent references and I put them there not because of attaching any particular value to them. It is a way of getting into what is truly an

enormous literature. What I have been asked to talk about is a huge job.

What I intend to do is talk about some of the problems and general issues that arise in designing media for the culture of embryos.

[Slide.]

Just very quickly, to remind you of what is happening, there are a whole variety of different ways of culturing embryos but a very common one is the droplet method that was first described by Gwalkin back in 1962 in my lab where you put droplets of medium on a Petri dish and then overlay that with mineral oil and the droplets remain intact there. Then, with a pipette, you can put embryos into the droplet.

[Slide.]

Then they are put into an incubator through which a gas mixture flows.

[Slide.]

Now, to talk about this in general terms, you can abstract the whole of that equipment. What we are concerned with here is the droplet in which the embryos exist surrounded by the medium. Then, beyond the mineral

oil, above it, is the gas phase. What we are concerned with physiologically are the rates of exchange of all the different compounds between the medium and the embryo and between the gas phase and the medium.

You can culture embryos in a system like this. The period for which they are cultured varies with the species. In the human, it is usually up to five days during which time, if you have got the right conditions, blastocysts will form.

From now on, I am going to concentrate of the problems of developing media. This is done for two reasons, to improve the yield of embryos for transfer back to the patients and the second reason is to design a mixture of a solution, or a medium, rather, that has minimum side effects.

[Slide.]

I would like to begin by some definitions. In tissue culture, one talks about different sorts of media. biological media are obviously media made up from natural biological fluids like serum or embryo extracts of various kinds. Those media we know nothing at all about their chemical composition.

Nowadays, people tend to use, or almost always use, chemically defined media. This is a very old idea. The first tissue-culture experiments ever done were done at Yale in 1907 culturing nerve tissue. Four years later, at the University of Pennsylvania, Margaret and Warren Lewis, who were parents of cell biology in this country, suggested that chemically defined media were the way to go.

Various media were developed. This subject really didn't get off the ground until about 1947 from work in Germany and England and also the United States. Very recently, because of the concern of natural proteins, we now read about protein-free media. Two other words that are frequently used which are very arbitrary; often we talk about simple media containing less than or equal to twelve components and complex media greater than twelve components. That is extremely arbitrary. You could use any number you want there but it is frequently used in the literature.

The Lewises published three papers, two in 1911 and one in 1912. I'm sorry; at that time, they were at Johns Hopkins University. I made a mistake. These media can be easily reproduced at different times and in

different laboratories. They can be varied in a controlled manner by selecting compounds and their concentrations.

They are free of unknown enzyme activities, and I added this in here to make it more modern, hormones and growth factors which may interfere with responses being studied.

There are two points I want to make about this. One is, these reasons are pragmatic. It is largely done to get repeatability between laboratories. The other thing is the composition of a defined medium is the composition when you start to use it.

Some people get confused and they say, well, it is not defined after you have incubated because it has been changed by the tissues you culture. That is missing the point. It is a known composition to start and experiment which everybody can do.

[Slide.]

Just a quick history on this. In the 1970s, Bob Edwards and his colleagues in Cambridge including Barry Bavister, who is here, started trying to develop media for culturing human embryos. In 1980, after the first test-tube baby was born, Edwards wrote this article in Science,

I think it was, human ova will tolerate a wide range of culture media. The most suitable and simple medium for fertilization is Earle's medium supplemented with pyruvate and inactivated human serum.

This is, therefore, a biological medium because, although this is a chemically defined one, you have added an unknown quantity in the serum. The same medium can be used for cleavage with higher concentrations of serum. They did get blastocysts from newly fertilized human eggs in this system.

The interesting thing is, since we know a lot about Ham's F10 is that Edwards said, in those days, Ham's F10 is unnecessary. What he was meaning is that this is a simple medium and this is a complex one with about thirty components.

[Slide.]

The development of chemically defined media started in the 1980s and the first description was by Menezo and his colleagues in 1984 who developed a medium that is widely known as B2. But if you go to the original paper, as I understand it, B2 was something developed for

the cow and B3 was a modification for the human. But, nowadays, B2 is generally used and it causes confusion.

Then, a year later, Quinn introduced what he called human tubal fluid which was a simple medium that many of you have used.

[Slide.]

One of the problems with those earlier media is that embryos only developed up to about the eight-cell stage. So the yield of blastocysts was very small. So a time came when people tried to make systems where you increase the yield of blastocysts.

In 1990, Menezo and his colleagues introduced the technique of coculture in which embryos are grown on a sheet of other living cells. Of course, that, then, immediately makes this a biological media system. Nowadays, it is not used very much and there has been concern with this over virus contamination which may be passed on to the embryos.

In 1995, Quinn modified his medium to make it in which glucose and phosphate was omitted. This was based on work that started with Barry Bavister in hamsters which showed that glucose and phosphate was inhibitory. This has

seemed to apply to many other animals, but we now know that this is not universally true. You can get media where glucose and phosphate are not toxic.

Then we have, more recently, the introduction of two-step culture methods pioneered by Gardner and Lane where you culture up in one medium for the first three days and then you switch to another medium. This has become very popular. There are about ten companies now manufacturing media for use in a two-step procedure. It has essentially become dogma, which I think needs to be challenged and I will talk about that a little later.

[Slide.]

When you think about the compounds you could put in a medium, there are the obvious ones that are oviduct-fluid constituents. There are ones that are found throughout the body, the second circle here, and then this is a set of every chemical that is available.

The mixtures that were usually used have practically no uniquely defined oviductal constituents. These things are now being described but they are not routinely used in media. What we are using is a solution

that is mainly made up of chemicals found throughout the body plus a few artificial ones.

[Slide.]

This is a medium that Joe Leitz and I designed in '89 called KSOM. You can see it is a simple medium containing various inorganic salts, lactate, pyruvate, glucose and glutamine. But the foreign substance that you never find in nature was EDTA.

So we had these different types of substances that are put into media. KSOM--this was the original KSOM which was good for going through what we used to call the two-cell block. It happened to be good for culturing blastocysts in many strains, but, as a result of work that was done in the Culture Club, this medium is now supplemented with amino acids and proves to be extremely effective in a whole variety of species, and I will be talking about that again a little later.

[Slide.]

The strategies for the design of media; there have been two approaches, one which I tend to favor and I suppose have provided more than anybody else is "let the embryos choose" strategy. This is really a bioassay. What

you do is you expose the embryos to a mixture of different concentrations of substances and see what they do, and you choose the mixture that seems to give the best result. It is not based on any particular knowledge of physiology.

The "back to nature" strategy, which was introduced by Henry Leese, who is going to speak tomorrow, attempts to make media in which the concentration of its components are those that you find in the oviduct, which makes intuitive sense. There are problems with applying this, however. We know the concentrations of only a very few compounds in the oviduct that you might want to use. It is also very difficult to collect oviduct fluid, to do analyses that you might believe in. There is a big literature on this but most of the analyses are questionable.

The only ones are the ones that we have used to study inorganic salts and Henry Leese has used to study carbohydrates where micropuncture techniques are used where you just put a needle in the top of the oviduct around the eggs and suck out a few nanoliters of fluid which, with microchemical elements, you could analyze. Probably, in

the end, both of these approaches will contribute to the design of adequate media.

I just want to point out the complexity of doing an assay. This is based on the results we got where we varied the concentrations of so-called nonessential amino acids and essential amino acids in different proportions. This gave a grid, a whole set of things in a factorial experiment. We measured the proportion of blastocysts that hatched and then, from that data, fitted this two-dimensional curve or surface.

This enables me to point out a very important fact in all of this. Supposing we fix the nonessential concentrations here and draw a line through here. The maximum we would find would be about this point. But the true maximum is somewhere up here. You would only find that if you happen to arbitrarily choose this point of non-amino acids.

What you have to do with this is to vary all these things simultaneously and see which is the best in order to determine the interactions. This is a prodigious task. If you have got twelve compounds in a medium, you will have to make up--you use the meter-2 concentrations--

you would have to mix up several thousand different mixtures to compare, which obviously you can't do.

There is a technique that was used to optimize processes in engineering, in industry chemical processes, called sequential simplex optimization where you model your mixtures not in two-dimensional space here but in multidimensional space and then do your experiments and use the data and analyze it to find a way to the maximum of this surface.

This is how we designed medium KSOM. It took us two years to do it, but it did result in a pretty good medium.

That sort of gives you a feeling of the problems involved in putting these mixtures together. In general, people in clinics are not interested in doing this sort of thing. It is very tedious and hard work making up all these solutions. So there has always been the tendency to buy off-the-shelf media that are commercially available.

Initially, this was a media that had been developed usually for the culture of human cell lines, cancer cells, and so on and were not tailored in any way to

the requirements for embryos. But they seem to work and Ham's F10 is one of these.

People are still using those media. Others were developed, as I mentioned, chemically defined specifically, the Menezo and the Quinn media. They are used quite widely. Now the two-step procedures have resulted in several different commercially available media.

[Slide.]

Now, having said all that, the first point I want to make is that it is inevitable that the embryos are stressed. All these solutions are artificial and there is no way in which we can make up a mixture that exactly imitates the condition in the oviduct.

If you depart from that, then the embryos, if the stress is too big, they obviously will die. It if is moderate, their adaptive mechanism is going to respond to it. So adaptation of embryos in culture, I think, is a very important subject which has not been widely studied. It has been discussed by myself and also Henry Leese.

All we can do, then, is to optimize the composition of the media by such techniques as bioassay.

[Slide.]

Now I want to talk about possible stresses that you might encounter. If you put a toxic substance in a medium, obviously, it is going to kill the embryos or retard their development. This is a totally unacceptable effect. Glutamine has been a substance of interest because it is in nearly all media, even in the early cell-culture media. One of the problems with glutamine is it is not stable and it breaks down into two toxic products if you store it in vitro.

This has caused trouble. If you have too high a concentration in the medium, then you can get toxic effects. Gardner and Lane in 1995 reported that glutamine in culture media for mice resulted in a significant level of exencephaly.

By reducing the concentration of glutamine down to low levels, you can reduce this and, in fact, we have done these experiments. It is a matter of what concentration you use.

The other alternative that is used by cell-culture people for years and is now beginning to be used in the embryo-culture media is to replace the glutamine with a dipeptide like glycyglutamine. This can get into cells

and, presumably, break down in the cells to give the glutamine which is metabolized so it doesn't have the opportunity to break down.

So this is one type of effect that you can get in culture, a toxic effect that can be often ameliorated by just reducing concentrations. Not always.

[Slide.]

There are two types of cells and embryos in culture, or even in the body, are always being subjected to. Metabolites are being produced which change the concentrations in the media and this is going to increase the osmotic pressure surrounding the embryos.

Other substances are produced that are going to change the pH in the medium, acidic metabolic processes. All cells are able to compensate for these changes in their environment by what are called homeostatic mechanisms. This certainly goes on in preimplantation embryos.

I will just try to quickly explain what is called regulatory volume increase and regulatory volume decrease. I will talk about the increase here.

If you put a cell in a hypertonic solution, water will immediately leave the cell and it will shrink. But

the cells have mechanisms to compensate for this change. This is a very fast reaction. A somewhat slower reaction is for ions like potassium and chlorine to move across the cell membrane into the cell. That will tend to draw water back into the cell and so cause it to swell up to its normal size.

The problem about moving ions into a cell is if they go up too high, the concentration becomes toxic. They start denaturing proteins and the cells will rapidly die. So, in evolution, what has evolved is the use of what are called osmotic osmolytes, various amino acids like glycine are osmotic osmolytes, so what the embryo will do in order to compensate against osmotic increase in the environment is to also move in the available organic osmolytes in the oviduct fluid.

This study is ow being extensively studied. Jay Baltz at the University of Ottawa has published many papers in this field and on the mechanisms that are involved in this transport. If there is very prolonged stress, this hypertonic solution is not relieved. In some cells, you can get a slow reaction. This has not been demonstrated in embryos probably because the time is never long enough.

These compounds start being synthesized in the cell. It could be sugars like sorbitol and so on. And there are genes involved in this. So prolonged stress will activate genes to make the needed compounds inside.

More recently, Baltz has also shown that genes probably are turned on to increase the production of the transport proteins that are in the cell membrane that are necessary to shift, say, glycine into the cell. This is a rapidly expanding subject on the physiology of preimplantation embryos which is very useful interpreting what is happening in culture.

This diagram here merely shows the reverse of putting--when you put these cells in hypertonic solution, which I won't go into.

So these are natural defense mechanisms against natural stresses that arise. They are fast. They immediately compensate and the embryos or cells survive quite--there is no problem.

[Slide.]

We are also discovering other defense mechanisms which may be against more serious threats. Again, this is a subject that is ten years old but we are still a long way

from understanding all this. But, in the body, there are proteins called heat-shock proteins such as heat-shock protein No. 70.

This is a very misleading name. It is due to the history in which these things were discovered because heat was used as stress. They are produced in response to all kinds of stresses, osmotic stress, poisonous chemicals and so on. They are a part of a group of compounds called molecular chaperons. Molecular chaperons are used to maintain the three-dimensional integrity of proteins in cells.

If under stress, when we say proteins get denaturated, they lose their three-dimensional shape and then they can't function properly. So these substances are important to protect embryos.

The appearance of this in development of the mouse embryo has been studied and also the cow. There seems to be what is called constitutive synthesis; that is, these are made anyway without any stress and it is not until about the four- to eight-cell stage that you can induce the production of this by exposing the embryos to stress. All of this work was largely done by pioneering

work in France. I can't remember all the authors offhand, but Professor Reynard is one of them.

So that is what happens. These are turned on due to activation of genes as well.

[Slide.]

To show the sort of thing that they can do, an experiment was done down at the Environmental Protection Agency by Dix and his colleagues. They cultured embryos in KSOM, not with amino acids. They exposed embryos to a certain concentration of arsenite. So you are trying to poison these cells with arsenite.

With a low dose, we find that the percentage of blastocysts that was produced was about the same whether arsenite was there or not. But if you introduced into the system this coated compound that is an antisense oligonucleotide that blocks the action of Hsp70, you can see the development, under these conditions, is severely affected. Even without arsenite, there is some depression presumably due to the effects of the arsenite.

If you knock out the HSP70, the number of blastocysts almost goes to zero. So here we can see that, under these conditions, the embryo can tolerate this dose

of arsenite but, if this substance is not working, they can't tolerate it. So there is a stress there and this is another mechanism that cells can protect themselves.

This sort of thing is probably going on more than we realize so that this needs more work done on it.

[Slide.]

The next slide is one of Christians. This is from Reynard's lab in France published in '95 which hasn't received too much attention and I think is very interesting. Here, they measured the heat-shock Protein 70 in mouse embryos growing in vivo under natural conditions. You can see, round about the two-cell stage, there is a slight peak there.

The two-cell stage is a very active stage of early development when the genes and the sperm are being turned on. This is what is called the zygotic transition where, before the zygotic transition, development is controlled only by maternal genes. Once the sperm genes are turned on at the two-cell stage in the mouse, then we have got a completely diploid set of genes available.

In the human, that transformation takes place at about the four-cell stage.

Here, they cultured mouse embryos in Medium M16. That is a medium that David Whittingham developed many years ago and which was used, and is used, extensively. It is a medium that is recommended in the standard manual on this field, Hogan's textbook. There are better media now but it was a good medium in its time.

You can see that the amount of stress this HSP70 is markedly increased around the two-cell stage, above normal. Whether this is occurring in other systems would be very interesting to know.

[Slide.]

Now I want to come to a subject which you have probably heard more about, genetic examples of stress. The first indication that media might affect gene action came from the studies of Ho, Eppig and Richard Schultz in 1995. They show that if you added--they were the first to actually add amino acids to our KSOM and show that it gave a much better development of zygotes to blastocysts.

The first medium that was ever used to culture embryo was Whitten's medium. This was in 1958. It is a simple medium. It was used for a long time until actually the M16 replaced it.

What they did was to compare the development of mouse embryos in Whitten's medium and this amino-acid-embellished KSOM and look at about eight genes. Half of the genes are what we call housekeeping genes that regulate the metabolic processes in cells. The other half were genes concerned with development.

What they found was that, in Whitten's medium, the housekeeping genes were turned on and worked as expected. The development ones were not. In the KSOM with amino acids, both the housekeeping and the developmental genes were turned on. So this was the first demonstration that genes can be affected by your culture conditions, whether they are turned on or not.

Then, in Schultz's lab at the University of Pennsylvania, an interesting discovery was made about genes concerned with imprinting. What was shown here, using again Whitten's medium in our KSOM.

[Slide.]

H19 is a gene that is concerned with imprinting and it is only expressed from the maternal allele. It is not expressed from the genes that come from the male. If you culture in Whitten's medium, there is abnormal

expression because the genes from both the male and female are expressed, which is not the normal thing, whereas, when you use this KSOM amino-acid mixture, the correct response occurred; only the maternal genes were turned on.

There are several other papers which I don't have time to go through in detail giving other examples where genes are definitely affected by media.

To sort of summarize about these adverse effects, although we can demonstrate these things, the big question is do they matter, are you still going to get normal babies despite this. The evidence that is available so far suggests that they really don't matter all that much. Normal babies were being produced when we assume that, with the media being used, these sorts of things are going on.

I think the reason for this is that, in development, there is a huge backup system, that if one system is not working, another system will replace it. This is speculation. I don't have any proof of this but this is an area that certainly would be very interesting to study at a basic level.

I would like to say just a few things about the two-step method of culture introduced by Gardner and Lane.

This took origin for two reasons. One was in order to overcome the toxic effects of glutamine, which tend to operate early on in development, and also the fact that analyses have been made by Henry Leese and Gardner in which we knew the concentrations of glucose and pyruvate and lactate.

We know that initially the glucose is low and then, at the time blastocysts form, the glucose is high. So you might want to use a medium that is low glucose to begin with and then switch to a high glucose.

This makes intuitive sense but there are exceptions where imitating the natural conditions don't work. Potassium is one of these. The more general things like osmolarity embryos develop naturally. According to Baltz, at a osmolarity of about 297 milliosmoles. The optimum for culture is about 270 milliosmoles, much lower. So we find the optimum is less than what you find in nature.

What Catherine Racowsky, who is going to speak next, and I have done is to use--we were interested in seeing whether KSOM was any good for culturing humans. What we found was that KSOM alone, from the zygote state to

the blastocyst, would give yields of blastocysts equal to the two-step culture method used in routine use in the clinic at the Brigham Hospital.

[Slide.]

These are human blastocysts. This was produced in five days with a two-step culture. At that time, the optimum two-step medium was not a commercially available one, but the medium P1 developed by Poole for the first three days, a simple medium followed by CCM, one of the in vitro life media.

That was working very well in the clinic in the Brigham. This is a typical blastocyst.

This is a blastocyst with the inner-cell mass here which was produced entirely from the zygote in five days without changing the media. We have done this many times and this occurs in high yield. Right now, in the process, we are testing how this translates into babies. We have several babies born using this system, but it is not enough to prove one way or another whether it is just as good as the two-step procedure.

This means that you may not have to go through the elaborate business of subculture in the clinic if this

is true. Also, when you renew a medium in the middle of this whole process, if there is a stress from the first stage, there is going to be a stress when you put it in the second stage so you might eliminate that.

This is a subject that is open for debate and you will see progress made, I hope, in the next few years on it.

I have stressed through all of this that concentration of substances is just as important as what you put in the medium. It is critical to know what the concentration is in order to critically analyze what is going on with these culture media, what is going on with the embryos.

Now, in my view, a very unfortunate development has been occurring in recent years. There are several companies that are now selling two-step media. They will tell you what is in the medium but they keep the concentration secret. So it is impossible to really do a scientific analysis of what is going on without that knowledge. They obviously do it for commercial reasons.

I think it is ethically wrong for doctors to expose human cells including embryos to solutions whose

composition is not exactly know, and by that I mean what is in the medium and what the concentrations are, which I have stated and published about before.

This thrives in secrecy. It retards development of better media, in my view. Through the advertising that is taking place of these media, you get the impression in the journals that these are the best media possible, so their use has become dogma.

Our work that I just described here with KSOM is suggesting that this should be questioned. This, then, I think is a very important ethical question for the group to consider whether we should use media of unknown composition.

I would add that the American Tissue Culture Association, back in the 1960s when chemically defined media for culture of human cell lines was being developed, a committee was set up to formulate the different media and what their compositions were exactly. So, when somebody said they used Ham's F10, they knew exactly what was in that medium.

The companies that manufacture those media and, to this day, have always adhered to those rules established

by the American Tissue Culture Association. It seems to me this sort of thing needs to be done in the assisted reproductive field.

So, in this talk, I have covered a lot of things. I have tried to give you a general feel that the development of media is a complicated matter. We don't have the ideal one. We will never get a complete imitation of the natural conditions and so there is still room for more work in this field.

Thank you.

DR. GIUDICE: Thank you, Professor Biggers.

Our next speaker is Dr. Catherine Racowsky who is Associate Professor of OB-GYN at Harvard Medical School and Director of the ART Lab at Brigham and Women's Hospital. She will talk on the Clinical Practice of ART.

Clinical Practice in ART

DR. RACOWSKY: Thank you very much. I would like to thank Phyllis Lepperd for inviting me to participate in this workshop. It is a real pleasure to be here and, clearly, with the multidisciplinary approach that we have, hopefully great progress will be made towards establishing

the guidelines and regulations that this field so desperately needs.

[Slide.]

What I have charged to do is give you a presentation on the clinical practice of ART.

[Slide.]

To do this, I am going to spend a little bit of time on a general background for those of you in the audience who don't routinely think about these issues from a lab prospective. I am then going to move to a brief consideration of controlled ovarian stimulation for ART, consider the issue of embryonic wastage and high-order multiple pregnancies arising from IVF and ICSI and then talk with you about strategies for embryo selection as we decide how many embryos and what quality embryos should be returned to patients, and then consider with you policies restricting the number of embryos to transfer and then, finally, to try to summarize all this for you in a brief forty minutes or so.

[Slide.]

To start, then, with just the consideration of the developmental time line of the human preimplantation

embryo, somewhere around 14 to 18 hours after exposure of the oocyte to sperm, the zygote, if, indeed, has formed, should exhibit the two spherical structures that are known as the pronuclei and then, several days following this, there is cleavage first to the two-cell stage, four-cell stage.

On Day 3, we believe the human embryo should be at the eight-cell stage and, if all is going well developmentally, the embryo then should continue to develop to form a blastocyst around Day 5 with implantation occurring around Day 6 and a half in the human.

[Slide.]

As we probably all know in this room, the first IVF baby was delivered in June of 1978, as John Biggers said, from Cambridge in England. This delivery arose from the transfer of an embryo on Day 2. It was certainly not a trivial task. Barry Bavister was very much involved in the development of this and it is my understanding that at least 100 attempts had to occur before the first success with respect to the actual delivery of a baby.

Natural cycles were used in those days in order to obtain oocytes, initially through laparotomy and then subsequently through laparoscopy.

There was very low efficiency of this procedure, obviously, and this can be considered to be due, first of all, to the availability of only one or two oocytes because these were natural-cycle stimulations and also with respect to suboptimal culture conditions. But, of course, there may be a large number of other reasons why the efficiency was so low.

[Slide.]

I raise for you these issues here; a possibility of an inadequate or unreceptive endometrium, asynchrony between the endometrium and embryonic development, sperm quality issues as published by a large variety of investigators looking at both cytoplasmic competency--we heard early regarding the contribution of the centrioles, indeed, nuclear problems within the sperm and, also, and very importantly, the possibility of oocyte quality and how this might really impact upon the very low efficiency of reproduction in the human.

[Slide.]

We know from a large body of literature that the delivery rate, and also the implantation rate, from ART procedures declines dramatically with maternal age. You can see here that this decline is particularly so after the age of around 39 with very, very low success rates ensuing in patients 40 years and older.

[Slide.]

There are several requirements for an oocyte to be able to be fertilized and to support normal developmental competency. I show you here what these requirements are. First of all, the oocyte has to be mature. Gerry Schatten talked today earlier regarding the omission of the polar bodies and the importance of this to compensate for the fact that two diploid genomes, if they came together, would give rise to a genetically out-of-control individual so you have to have two haploid cells to start with.

The oocyte, known as the mature oocyte, has to have completed a very, very complex series of events called cytoplasmic maturation so that it can complete meiosis, finish the second meiotic division, it can support a pronuclear formation, syngamy--the joining together of the

two pronuclei--and then, indeed, activation of the embryonic genome which, as John Biggers indicated, occurs somewhere around probably a little bit later than four-cell--somewhere between the four- and the eight-cell stage in the human.

But, in addition to cytoplasmic maturation, the oocyte also has had to complete nuclear maturation. A normal oocyte completing nuclear maturation should be euploid. In the human, it should have 23 chromosomes. This process is a very complicated process, a very delicate process and a lot of errors are made during this process giving rise to what we call an aneuploid egg, one that has either too many chromosomes or too few chromosomes.

[Slide.]

Indeed, if one looks at the literature, and Sante Imuni from St. Barnabas Hospital in New Jersey has been a major contributor to this--if one looks at the literature, one sees that the incidence of aneuploidy, not surprisingly with respect to the slide I showed you earlier showing the decline in delivery rates, shows that the incidence of aneuploidy increases dramatically with maternal age.

Corresponding with this, since the vast majority of aneuploidies are lethal, implantation decreases with maternal age.

You notice here the percentage of the total oocytes examined, around 40 percent aneuploidy oocytes at these elevated maternal ages. However, I will say that there is other literature which shows that this number is actually considerably higher. On the order of 60 to 70 percent of human oocytes in these ladies of advanced maternal age are, indeed, aneuploidy.

[Slide.]

If you consider for a moment the natural fecundity of humans, it is very low. It is incredibly low compared with other species. The data here shows you a table drawn from Leridon's publication in 1973 showing the incidence of survival of eggs during gestation. You can see that there is a very high incidence of death of oocytes or embryos early on so that, from the weeks post-ovulation 0 to 2, of the order of around 50 percent of embryos are lost before any pregnancies really become properly established.

So there is a very high attrition of human embryos during the natural setting and I want us all to bear this in mind as we think through the incidence of embryonic wastage from assisted reproductive technologies.

[Slide.]

So, in light of the fact that we have said that efficiency is very low if you use a nonstimulated cycle, as shown by all the work from Steptoe and Edwards way back in the 70s and early 80s, in addition to the fact that there are an awful lot of errors made in human oocytes during the normal process of meiotic maturation, we have, then, to increase the number of oocytes in order to increase the likelihood of success.

Controlled ovarian stimulation is the course designed with this goal in mind in order to stimulate a controlled growth of a follicular cohort to obtain a cohort of oocytes that have developmental competency, a large number, so that we end up with more embryos than we actually need to perform a fresh transfer but then the ultimate goal being to actually improve the success rates of in vitro fertilization.

[Slide.]

I put this slide up to remind us of what actually we are doing when we perform controlled ovarian stimulation. In a normal menstrual cycle, there is cohort of growing follicles which actually have been recruited many days prior in previous cycles, but there is a cohort of growing follicles from which a dominant follicle is selected. Usually, it is only one dominant follicle.

The remaining follicles undergo a process of degeneration or atresia and get lost from the cohort quite early on during the menstrual cycle, around Day 3 to 5. When we apply exogenous FSH in a controlled ovarian-stimulation cycle, what we are doing is we are rescuing these follicles that would otherwise undergo degeneration and atresia and bringing them back on the pathway of follicle development and growth.

So we must remember this when we starting thinking about the quality of the oocytes we are retrieving from our patients and the quality of the embryos that arise and the consequence of this policy being disparate in many respects giving rise to a high incidence of embryonic wastage.

[Slide.]

The goal with controlled ovarian stimulation is, as I have alluded to, to increase the number of meiotic immature oocytes. But we know of work from Wynn and many other people that not all oocytes that are retrieved from a controlled ovarian stimulation cycle are meiotically mature. Actually, the proportion is of the order of 80 percent.

[Slide.]

Not all of these meiotically mature oocytes fertilize normally. In a well-run ART lab, you can expect somewhere between 80 and 90 percent of mature oocytes to fertilize under normal circumstances. But then, of those that do fertilize, not all morphologically normal embryos have the ability to make a baby. There are a lot of embryos that appear morphologically abnormal.

[Slide.]

This slide has been put together from four embryos that arose from the same cohort from one patient and one stimulation cycle. You can see the enormous disparity in quality here. I want to just take a second for those of you who don't think about grading embryos and

are not used to it, just to briefly describe to you how we grade embryos in the clinic on a day-to-day basis.

The first and foremost thing we look for is cell number. We ask the question, how many easily observable, readily identifiable, cells are present. We also look for the extent of fragmentation in the embryo. As you can see here, one of the cells has actually exploded to form a large number of cytoplasmic fragments.

There has been some work to show that there is nuclear material in some of the fragments. Other work has shown these fragments to be enucleate. We really don't understand why fragmentation occurs in the human, although I will hasten to say that, at least at low level, it may be beneficial and some way in which the embryo undergoes some self-corrective mechanisms.

So we look at cell number. We look at the extent of fragmentation and we look at the symmetry of the blastomeres. Here you can see a much larger blastomere and here a much smaller one. So I am not really referring to how symmetrical the overall embryo is.

Here you can see what we call in the lab sausages. But you can see we are not really talking about

symmetry of the whole embryo but we are talking about how symmetrical each one of the cells is with respect to the other cell. I am going to be saying more to you about the relationship of these sort of what we call three first-line parameters for assessing embryo quality in just a few moments. There are other ways that we also look when we try to evaluate the embryos, as I will be discussing.

[Slide.]

So, because of this disparity, then, in embryo quality from a technical cohort of embryos, there inevitably is going to be embryonic wastage. What I do here is to create a scenario for you where about ten mature oocytes are retried, on average, in this country per ten patients giving rise to 100 mature eggs.

As I have said, one should expect around an 80 percent fertilization rate from those hundred eggs to give you eighty embryos. So, already, we have wasted twenty of those oocytes.

If one is performing a Day-3 transfer, in this country, there is very much a movement towards reducing the number of embryos and I am going to talk with you about that, but let's, for the sake of argument, say three to

four embryos are transferred on Day 3 giving rise--if we take four embryos as the example, that would give rise to forty embryos being transferred.

Some of them, that were suitable for transfer, would remain, that might be suitable for transfer in a subsequent cycle and would end up being frozen. We can assume, perhaps, five of our patients had freezing with two embryos being frozen giving you and additional ten embryos.

So forty plus ten is fifty embryos from the eighty so we have now lost another thirty embryos during these procedures. Of those embryos that are transferred, of course not all of them implant. We can expect around a 20 percent implantation rate from Day-3 transfers.

So, of our embryos, the forty embryos that were transferred with a 20 percent implantation rate, one ends up, then--actually, thirty embryos transferred--one ends up with only six embryos implanting. So, again, there is an attrition of embryos.

The point to be made from this slide, then, about 75 percent of mature oocytes and around 70 percent of embryos arising from ART, are actually wasted. So this is an incredibly wasteful procedure a large part of which is

undoubtedly due to inherent problems within the oocyte but, undoubtedly, also, is contributed by our inadequacy of the culture conditions as discussed by John Biggers.

[Slide.]

So the dilemma, then, with cleavage-stage embryo transfers is the obviously following: not all human embryos, as I said, develop normally and so we have to ask a question, what is the appropriate number of embryos to transfer into every single patient that walks into our IVF clinic.

We want to maximize the chance of every single patient having a pregnancy while minimizing the risk of a high-order multiple pregnancy. I am going to focus particularly on triplet gestations and above although I am sure obstetricians in the audience would also argue that twin gestations are problematic.

The upshot of all this, then, is that, on balance, too many cleavage-stage embryos continue to be transferred from Day-3 transfers in our country.

[Slide.]

This slide shows you the data originally put together by SART and now, since 1995, published by the

Center for Disease Control, showing the national delivery rate for embryo transfer in blue and, in red, the percentage of pregnancies that actually had more than two fetuses.

Interestingly, in '95 and '96, you can see that overall pregnancy rates were relatively low. Particularly in 1995, the triplet rate, triplets and above rate, was extremely low, undoubtedly not so much due to too many embryos being transferred back but probably inadequacies of the culture media affecting the developmental expression of the oocytes and, therefore, reducing overall implantation rates.

But, as we go better at culturing embryos and as there was a further sort of push to improve pregnancy rates because of interprogram competition and real pressures from the patients, we became much more aggressive in the number of embryos to transfer. Indeed, in 1997, you can see that the national rate of high-order multiple pregnancies--that is, triplets and above--was of the order of 15 percent.

We then started, as a field, to critically question what we were doing. You can see that, since 1997, there has been a gradual trend downwards with respect to

the triplet and above pregnancy rate with, nevertheless, a continued increase in the overall pregnancy rate. So this is reassuring.

You will notice that the most recent year that we have data for is 1999. The 2000 data will be released, as I understand it, in December of this year.

[Slide.]

So, because we have agreed that we generally make more embryos than we need to make in order to perform a transfer and we also know that there is enormous disparity in embryo quality, we have a responsibility to improve the methods with which we select the best embryos of a cohort for the transfer.

There are many different ways that we are doing this and there are new ways on the horizon. I list for you, first of all, the morphological characteristics. I talked with you about the three conventional variables we use, cell number, fragmentation and symmetry.

I am going to be discussing with you, in some detail, more newly identified morphological characteristics which are now coming on the horizon and being routinely applied by many clinics. I am going to address with you

briefly aneuploidy testing using FSH and how this can impact repeat IVF-failure patients, advanced-maternal-age patient and repeat-miscarriage patients.

I am going to talk to you a little bit about cell-selection approaches particularly with respect to extending the duration of culture from Day 3 to Day 5 so the embryos are challenged to adjust to this additional two days in culture after the activation of the embryonic genome to perform blastocyst transfers.

Finally, although I am not going to say anything about this today, the utility of noninvasive biomarkers which undoubtedly has enormous use in the field in the future and a lot of work is being done at the moment, including our own program at the Brigham, to try to identify markers that may have utility for selecting the best embryos available from a cohort.

[Slide.]

In order to really get a handle on the relevance of these parameters--cell number, fragmentation and symmetry--the best way to do this is to have a dataset where you are tracking individual embryos to babies, so you

know for sure that every single embryo that is being transferred has actually made a baby.

But you also need to know, if you do that, somewhere around 50 percent of pregnancies result in two babies being formed, gestations. You also need to control for the parameters appropriately

[Slide.]

What I want to do is just very briefly talk to you about a study that we finished and is now being prepared for publication looking at the impact of these variables and how you can actually start making definitive statements about the morphology of the embryo and how it affects a pregnancy being established.

So you can see you need a very large amount of data to do this. This data was drawn from our experience in 1998 through 2001 at the Brigham. You can see that nearly 3,000 embryo transfers were involved in this analysis initially of which we chose to select 803 Day-3 transfers in which the patients were youngish, less than or equal to 37-year-olds and on their first ART attempt, to try to control for some of the patient variables that might impact.

Then, if you think about it, there are various outcomes that can occur from a transfer. You can either have no pregnancy whatsoever. You can have a failed pregnancy. You can have what I refer to as nonequivalence; that is, a different number of fetuses developing from the number of embryos that go back but all embryos being of identical morphological criteria with respect to cell number, fragmentation and symmetry so that you know for a fact, if one baby has been formed, you know exactly what that embryo looked like at the time of transfer even if other embryos were transferred with it, albeit all of the same morphological appearance.

Then the other scenario would be the equivalence where you get the same number of babies being formed as embryos going back. So you can track exactly the embryos.

So, of this nearly 3,000 embryo transfers, we ended up with thirty-three embryo transfers of nonequivalence and 119 transfers involving what we refer to as the equivalence, involving 71 embryos being transferred here and 247 embryos being transferred in the equivalent group.

[Slide.]

What we found was the following with respect, first of all, cell number and how this impacts upon viability. I should define viability for you as a fetus at least at 12 weeks of gestation. On the X axis here is the number of cells in the embryos. As you can see, as one might expect from what I said earlier, we believe that a human embryo should be at the eight-cell stage on Day 3, you can see that a significantly higher viability rate occurred when the embryos were transferred to the eight-cell stage.

Very interestingly, however, the very fast-cleaving embryos, more than eight cells on Day 3, actually had a lower implantation rate than those that have eight cells. This has been a question that we have constantly grappled with at the bench in the IVF lab. If you are faced with trying to choose two embryos, say, from a cohort where there may be two eight-cell embryos and two ten-cell embryos, everything else being equal, which embryos do you pick out for transfer. This is the answer; you pick up the eight cells over the faster-cleaving embryos.

This ties up very nicely with some recent work of Sante Imuni who has looked at aneuploidy rates of embryos

with respect to cell number and has shown the aneuploidy rates to be higher in the faster-cleaving embryos. So this, I think, from a cytologically point of view, also makes some sense.

Looking now at the middle panel, the fragmentation, as I alluded to earlier, embryos that have few fragments may be just as viable as those with no fragments and, indeed, the data bears this up, no significant difference in viability rate if the embryo has up to 10 percent of its volume fragmented as compared to no fragmentation.

But you can see that there is a very dramatic decrease in viability as fragmentation level increases above the 10 percent mark.

Finally, with respect to the asymmetry, how symmetrical each blastomere appears to all other blastomeres in the embryo, you can see that when there is no asymmetry, viability is higher than when there is moderate asymmetry and when there is severe asymmetry where viability, in this dataset, at least, was extremely low.

[Slide.]

With these types of analyses, then, we gather a paradigm that we now use at the bench in the lab. We are actually going to reanalyze this data to learn more about interactions of the three different characteristics. But we have now this policy in the lab which tells the embryologist what order embryos should be picked up according to cell number, percent fragmentation and asymmetry.

Here, just to summarize what I have said, is the expected viability given these three parameters. You will notice that those embryos that have more than eight cells, even with no fragmentation whatsoever, should be picked up, selected for transfer after consideration has been given to eight-cell embryos with actually up to 25 percent fragmentation as long as there is no asymmetry.

[Slide.]

So, now, turning to strategies for improved embryo selection, thinking about more newly defined criteria, here I am going to address pronucleus scoring, early cleavage of embryos and early compaction.

With respect, first of all, to pronuclear scoring, and I should acknowledge Lynette Scott who is in

the audience who has really been at the forefront for pushing this as a way of evaluating early embryos. The work pretty much followed Testart's work that was published a few years earlier.

What Lynette has done, and she now has actually a more advance scoring system but it is based on the same sort of characteristics--what she has done is to ask questions as to how the alignment of the nucleoli within the two pronuclei actually impact upon developmental potential. She has shown that if an embryo has what she calls a Z1 or, in England, a zed 1, a Z1 or a Z2 score, the implantation rates are higher than those with Z3 or Z4 scores.

This panel on the right here shows you the number of embryos transferred having either a Z1 or a Z2 score of the total that were transferred. The blue column show the pregnancy rates and the red show the implantation rates. So that, as early as Day 1, at the fertilization check, fourteen to eighteen hours after insemination, you can start learning something about the developmental potential of the embryos based upon the polarity of the nucleoli in the pronuclei.

[Slide.]

More recently, Odeni Sakkas' group has looked critically at the question of early cleavage of embryos later in the day on Day 1 to ask whether, if one can identify early cleaving embryos, and this slide here shows you an embryo just about to undergo cleavage--here is the cleavage furrow here to enter the first mitotic division to form the two-cell stage.

What Odeni's group has asked is if, indeed, one can identify early cleavage embryos late on Day 1, is this some marker for developmental potential of either individual embryos or the whole cohort of embryos. His data has clearly shown that, according to the number of early cleavage-stage embryos which are transferred, one can expect differences in pregnancy rate, again shown in blue, and implantation rates show in red.

You can see that when there are no early-cleavage embryos evaluated and put back at the time of transfer, pregnancy rates are on the order of 25 percent whereas they go up really dramatically to 50 percent when there are a larger number of early-cleavage-stage embryos transferred.

So, again, on Day 1, one can learn a lot about the embryo in predicting the viability of the embryo and, indeed, for the practical perspective of selecting embryos for transfer.

[Slide.]

Now, this data here is concerned with early compaction of the embryo and this is work that has arisen from our group at the Brigham, Natalie Cekleniak--she graduated from her clinical fellowship two years ago and is very interested in embryo morphology and really spearheaded this work.

But she asked a question, is there any relationship between the incidence of early compaction of embryos on Day 3 and viability of the embryos after transfer and showed very clearly that, in embryos that have what we call in our system Grade 3 early embryos have a compaction score. But these embryos are the best embryos to pick up for transfer.

When I first went to the Brigham five years ago, this phenomenon wasn't called early compaction. It was actually called poor membrane definition, sort of with a negative connotation. The embryologists were really

avoiding trying to pick these embryos up for transfer. But it just goes to show how little we knew then and still we don't know a great deal more. We some more than we did then. Thank goodness I can say that.

But, nevertheless, the point to be made is that if you critically look at the embryos and think about the development of the embryos, you can learn so much from them just from a morphological point of view and continue to push to pregnancy rates up by careful evaluation and selection of the embryos for transfer.

[Slide.]

Now just to move to Day 5 transfers and to ask the question regarding the quality of blastocysts on Day 5. This was addressed earlier I believe by Gerry Schatten or it may have been by John Biggers considering a blastocyst is not just a blastocyst. You have to look very critically at the embryos. You have to understand what you are looking for with respect to cell number, disposition of the cells, the presence or absence, indeed, of the inner cell mass so critical, obviously, for forming an embryo. Otherwise, you get a blighted ovum.

You have to consider these things carefully. You can go back retrospectively and look at your data and ask the question, what is the implantation potential of embryos exhibiting particular morphological features on Day 5.

[Slide.]

These data here on the right show you our data from the Brigham showing that an expanding blastocyst and expanded--so here is an expanding blastocyst. You can see the zona pellucida hasn't completely thinned whereas here it is very thin at the expanded state.

But the implantation potential of expanding and expanded blastocysts is actually identical. So, if you think about the other characteristics that are important with respect to the number of cells in a cell mass, the completeness of the troph ectoderm and how the cells are disposed, one with respect to the other, that is going to be very important information when picking out the best embryo for transfer

[Slide.]

So then, with this brief discussion on morphological evaluations, I think it is fair to say that you can develop a sort of a pedigree for each individual

embryo for evaluating its developmental potential based on morphological appearance and use sort of this panel of characteristics that I have described for you to improve the implantation potential and the success of an ART cycle, ranging from the 2 PN pronucleate stage all the way through to the blastocyst stage.

[Slide.]

With respect to aneuploidy testing, and I am not going to say a great deal about this, but just simply to point out that there is evidence that using aneuploidy testing with FISH has, indeed, reduced trisomic pregnancies overall, in some studies has increased IVF implantation rates and reduced spontaneous abortion rates as shown by Sante Imuni. There are a large number of probes now commercially available for targeting specific chromosomes and many more that are up and coming on the horizon.

I would just like to say one thing about aneuploidy screening for advanced maternal age patients. There are several programs, both in this country and also worldwide, that are starting to apply this routinely for patients that are thirty-seven years and older. There is no real scientific evidence to support the efficacy of

this. By removing a cell from an embryo, you inevitably are going to compromise the developmental potential of that embryo.

So this is just a cautionary note for us, I think, to think about and possibly even discuss further really whether it is ethical to apply a screening test such as aneuploidy screening carte blanche across the board for patients that are getting on in years when we don't know what the proven efficacy is of it and we might actually be seriously reducing their chances of conception by applying that technology.

[Slide.]

So, with respect, then, to self-selection approaches, and we have heard a lot from Dr. Biggers about the culture conditions and how important it is not to challenge the embryos and not to force them to be put into stressful situations, the movement of blastocyst transfer very much came into its own in the mid-1998s, late 1998s. It wasn't a new procedure, as was discussed earlier by Dr. Biggers.

Early on in the field of IVF, human blastocysts were being made, but being made at very, very low

frequency, indeed. But, with the establishment of the two-step culture systems of David Gardner et al., there was a real move to establish Day-5 transfer as sort of the panacea of ART, that this was going to solve our problems for addressing the number of embryos to transfer back and putting back only those of high quality because, by taking the embryos from Day 3 to Day 5, you really are challenging the better quality ones to make the blastocysts.

So this seemed to make a great deal of sense. One could then get away with transferring only one or two viable embryos. Theoretically, triplet gestations should be eliminated under those circumstances and overall IVF success rates should go up as a result of that.

However, I will say that we have found that this procedure, with the technology as it is today, with the development of the culture systems, is applicable, but applicable in selected patients only and appears actually detrimental in some patients.

[Slide.]

The next slide shows you data that we published, I believe it was in the Year 2000 in Fertility and Sterility, where we asked the question with respect to the

number of embryos on Day 3 having eight cells, what is the pregnancy rate if the patient has a Day-3 transfer, shown in red, or a Day-5 transfer, as shown in blue.

What we found from this retrospective analysis was that, if a patient has no embryos on Day 3 that have at least eight cells and has a Day-3 transfer, she has a 33 percent chance of a pregnancy whereas, if that same patient is taken out to Day 5 with respect to the number o embryos having at least eight cells, a 0 percent chance of a pregnancy.

If you look at what we call the sort of moderate-quality cohort group where there were one or two embryos in the cohort having at least eight cells, pregnancy rates were identical, whether the transfer was performed on Day 3 or Day 5 and, likewise, if there were three or more embryos having at least eight cells.

So what this data showed us, or told us, was that the culture systems were not as good as the uterus at rescuing these ostensibly lower-quality embryos, those that were struggling, slurring cleavage, undergoing cleavage in culture. They needed to be got back into the uterus earlier and that you were going to severely compromise

their developmental potential if you continued to take them out for an additional two days in culture.

[Slide.]

So we became much more conservative with respect to those patients that we offered a Day-5 transfer to. These days, in 2002, only 15 percent of our patients at the Brigham actually have a Day-5 transfer and we are very stringent as to whom we offer a Day-5 transfer to. We want the cohort to have at least three embryos having at least eight cells.

We want to have at least eight embryos in the cohort and the reason for this is world experience shows it in an overall population only around 40 to 50 percent of embryos will make it to the blastocyst stage on average. So, in order to have a few blastocysts from which to select the better ones for transfer, mass tells you that you need to have at least eight embryos at the fertilization check to maximize that chance.

We only offer Day-5 transfers to patients that are under 40 years old. The reason for that is that aneuploidy rates, as I have indicated, increase dramatically in this age group, 40 and above. We can get

away with being very aggressive with respect to the number of embryos we transfer on Day 3 in this group and, actually, there are really rather few patients that have more embryos than we feel comfortable transferring back on Day 3 anyway, so what benefit is there, I might ask, in doing a Day-5 transfer under those circumstances.

So we are very conservative in the older population. We will do it if a patient wants it. We had as patient recently come back for her third child in our clinic, the second of whom was conceived from a Day-5 transfer and the patient is now 41 years old.

But because Tommy was born from a Day-5 transfer, she really wanted to have a Day-5 for her third cycle, and we are awaiting the pregnancy test. She actually made some nice blastocysts so, hopefully, it will be successful for her. But we do tend to be very conservative in offering Day-5 transfers to the older patients.

In addition to that, we don't use Day-5 transfers for egg-donor cycles. This is a big controversy in the field. There are some programs that argue vehemently that Day-5 transfers are appropriate for egg-donor patients because these tend to be younger patients. The egg quality

and the embryo quality tends to be much better and because this, at least ostensibly, should be the way to control the triplet pregnancy rate, it makes sense to do it.

We have found that our freezing rates with blastocysts are not as good as they are with Day-3 freezing. Egg-donation cycles are out-of-pocket for the patient, and the patients, generally speaking, are very interested in banking embryos as well as having a fresh transfer.

So our reasoning has been to maximize the cumulative pregnancy rate from any one single retrieval, that we will only take a patient who is having egg donation to Day 5 if she absolutely wants it.

All this is with the caveat that you know how many embryos to put back on Day 3 to control your triplet rate and I am going to be addressing that shortly with you.

Finally, we offer Day-5 transfers only to IVF patients and not to ICSI patients. This gets to Gerry's discussion earlier and to Dorrie Lamb's discussion of the quality of embryos from ICSI. We have found retrospectively looking back and comparing our pregnancy rates from Day-3 transfer ICSI patients and Day-5 transfer

ICSI patients, the pregnancy rates are significantly lower with the transfers on Day 5.

So, immediately, then, from this analysis, if one can assume that all variables have been controlled, and that is a really big if--I will be the first to admit it because of all the different variables that come into play--but if you can assume that all variables have been controlled, then one has to reason, then, that there is some compromise to the embryos arising from ICSI which results in these embryos being further challenged by taking them out for these last two days in culture to Day 5.

So it is a very rare event in our program that we will perform a Day-5 transfer in an ICSI cycle.

[Slide.]

So to get to the issue of the policies that restrict the number of cleavage embryos to transfer. This is the really sticky issue and the one that we are very much aware of and very much concerned with in the field today.

[Slide.]

In 1999, November of 1999, the SART Practice Committee published a report, guidelines for the number of

embryos to transfer. I stress the word "guidelines." These are not regulations as exist in England today and in other European countries such as Germany.

As you can see, on the left-hand side here, as one goes from the most-favorable-prognosis patient to the least-favorable-prognosis patient, one, according to these guidelines, can, with reason, increase the number of embryos to transfer so that, in patients that are over 40, according to these guidelines, we could put back five embryos or less.

In addition to publishing these recommended guidelines, the report stressed the following: individual programs are encouraged to generate and use their own data regarding patient characteristics and the numbers of embryos to transfer.

In other words, each program has a responsibility of going back into their own data, learning from their own data what works in their program to get a handle on the unacceptably high triplet rates which, in 1997, as I have indicated, were of the order of 15 percent by pregnancy in our country.

[Slide.]

Actually, it was a little bit before November of 1999, in January of 1999, at the Brigham, we set about doing this because, at that time, based on our data in 1998, we had a 12 percent triplet rate by pregnancy in our program.

Bob Barbieri, my Department Chair, came into my office in December--actually, it was Christmas and most of the faculty had gone for the holidays. He said, "Catherine, deal with the triplet rate in our program." I looked at him and I said, "Okay." So I spent most of my Christmas vacation in the data, wading around in the data, trying to understand what determinants might be useful to target as predictors of high-order multiple pregnancies and how, then, we might use those determinants to control our triplet rate, triplet-and-above rate in our program.

So I performed a retrospective analysis and I compared cycles giving rise to one or two fetuses with those giving rise to three fetuses or more. I proposed the following three what I call high-order multiple HOM determinants; patient age, the ART attempt number, and the number of Day-3 embryos having at least eight cells.

Using these three determinants, we then set forth to develop an algorithm. This algorithm is actually law now in our program. It has undergone six assessments. I am actually going to present data on five of them because we are just in the process of analyzing the efficacy of the sixth refinement that we have performed.

So, from January of 1999 to the present, we are still actively, proactively, working on trying to improve this algorithm in our program.

[Slide.]

Just to give you a feel of the sorts of things that we sort of developed as we got under way with this--so this was algorithm No. 1. This was for patients that were on their first through third ART attempt. You can see here the patient age and here the embryo cohort stratified with respect to those embryos having at least eight cells and those having three embryos or more with high cleavage stages.

We proposed the maximum number of embryos to transfer. You can see, not unreasonably, as patient age has gone up, so, also, could we go up to five embryos in patients that were over 40 years old. This was pretty much

following the guidelines that were published in the SART report although, as I say, we actually did it a little bit earlier than that report came out.

[Slide.]

Now to show you how we have sort of refined things. Now algorithm No. 5, and this just for the younger patients. You can see that we are much more discriminating now with respect to how the patients are stratified, to gauge the maximum number of embryos that is appropriate, remembering that all the while we are trying to maximize the chance of any pregnancy at all for a patient while reducing the risk of a triplet pregnancy to the minimum.

So you can see that, even in very young patients, if they are on their fourth ART attempt or more, in our program, and I should stress that, we only consider those attempts that have been performed with us because it just became an impossible undertaking to consider those from other programs because there were so many unknown variables. So, to try to keep things under control, we made the decision as a group that we would just consider those attempts that a patient has undergone in our program.

But you can see, in these very young patients, regardless of the number of embryos having at least eight cells on Day 3, we can put back as many as four embryos in this patient group which is really aggressive in a young patient population.

[Slide.]

This slide shows you the results of these algorithms to date. You can see, in 1998, as I have said, our triplet pregnancy rate was way too high, 11.8 percent. As we started introducing the algorithms and refining them, you can see how algorithm No. 2 and 3--I think No. 2, you can see that the overall pregnancy rates actually started to go down although we became quite effective in controlling our high-order multiple rate and there was a lot of unease amongst the group because pregnancy rates overall were going down and there is such pressure to keep them up all the time, of course.

So then we went back and looked at all the data again that we had achieved over the previous eight-month period between algorithm No. 2 and algorithm No. 3, and were able to continue to reduce the pregnancy rate but not markedly impact on the overall ongoing rate.

But, during the course of time, and now algorithm No. 5, and it looks like algorithm No. 6 is going to even be more effective, you can see that we have actually been able to overall increase our ongoing pregnancy rate across the board in the program and, nevertheless, pull our high-order multiple-pregnancy rate down.

It is now at 4.8 percent. It is not 0 percent. It is not the 2 percent that is reported in Germany or the 1.8 percent that is reported in the U.K. So we still have a lot more work to do in this regard but at least we feel that we have made some great inroads in addressing the problem and maintaining the overall pregnancy rate for all patients coming through our program.

[Slide.]

In closing, then, it is fair to say that humans have a low natural fecundity. That is a given. There is a high instance of oocyte anomalies that definitely increases with maternal age. There are great improvements to be made with respect to the controlled ovarian stimulation regimens in order to maximize oocyte yield and also, very importantly, of course, to maximize the developmental potential of each and every oocyte that is retrieved.

[Slide.]

There have been improvements in the culture conditions. A lot more work needs to be done to continue to create in vitro environments that better suit the physiological needs of the human oocytes and embryos. There are refinements in embryo-selection techniques which, coupled with the use of effective transfer policies, should continue to maximize the likelihood of a pregnancy while minimizing the risk of a high-order multiple pregnancy.

So, overall, there is a movement in the field to continue to increase pregnancy rates while, hopefully, continuing to decrease the triplet and above pregnancy rate.

I close, and I have never done this before--but I close by dedicating this lecture to my triplets, Adam, Lauren and Daniel, who will be sixteen years old in December.

Thank you very much.

DR. GIUDICE: I guess that is a real testimony from bench to bedside, or the other way. Thank you very much.

Before we open the session to further discussion, there are three individuals, I understand, who would like to make comments that are limited, I believe, to five minutes each. These include Dr. Barry Bavister from the University of Wisconsin, Dr. William Gibbons from Eastern Virginia Medical School and also Dr. Gabor Huszar from Yale University.

Dr. Bavister?

Public Comments

DR. BAVISTER: I would like to, first of all, thank the organizers of this meeting for allowing me to present a few minutes, to make some important points about evidence-based ART.

[Slide.]

First of all, I would like to establish my credentials for addressing this meeting because, later on, I am going to make some somewhat critical remarks and I want you to think I have at least some qualifications for doing this.

Basically, I have been a reproductive biologist for thirty years, card-carrying one, anyway. A lot of my work involves nonhuman primates, which is the point I want

to emphasize. I also want to emphasize that the NICHD has funded my research continuously for almost twenty-five years and I am very grateful for that.

I was a member of the notorious Culture Club, which has been mentioned several times already, and I have been a professor for about twenty-five years as well. I currently serve as President of the International Embryo Transfer Society which is an organization representing basic researchers in embryology and embryo-transfer practitioners from thirty-seven different countries.

[Slide.]

I was also privileged to work on the first documented human in vitro fertilization with Bob Edwards in 1968 and my lab in Wisconsin developed the first reliable protocols for nonhuman-primate IVP.

[Slide.]

Having said that, I want to add my five cents-worth to what Dr. Racowsky just so eloquently told us and that is that the efficiency of current human IVP in vitro production--I hate the word IVF--but IVP is not very efficient. According to the 1998 SART data, on a per-transfer basis, the clinical pregnancy rate is 35 percent

and this drops to 29 percent for live-baby rate which is a considerable loss.

But, if one assumes, and all the rest of the data are assumed, that three embryos are transferred in each procedure, and that is, obviously, an average, ballpark figure, then the CPR per embryo transferred is only 12 percent. If one looks at the number of embryos produced, let's suppose those are ten per cycle, then the CPR drops to 3.5 percent and so on.

The final efficiency of human IVP or ART, if you like, per oocyte retrieved and inseminated may be less than 5 percent. These hypothetical data ignore multiple pregnancies but you get the point, that there is an awful lot of wastage, as Dr. Racowsky pointed out, in human ART. The difference, I want to emphasize, between these very low efficiencies and this very high efficiency is solely due to the skill of the embryologists in the lab for picking out the best embryos.

But, since we really don't know too much about how to select the best embryos, we need to give the embryologist more tools for doing their job better and eventually raising this percentage.

[Slide.]

The current practice of clinical IVF or ART, in the United States, operates virtually in the absence of a base of animal embryology data for guidance and for verification of embryo protocols. The reason for this is that almost all embryo research supported by the U.S. government is relying on rodent models, primarily mice.

I do not want to say that this is bad science. It is just that we need more.

And, I'm sorry; some of these slides didn't get xeroxed.

However, there are major problems with using rodents as models for humans. Humans, of course, are primates as are Old World monkeys. I have listed some of them here. I don't have time to go through them. I would point out that, for example, a major difference is the centriolar inheritance. This work was largely done by Dr. Gerald Schatten. It would be quite inappropriate to use a rodent model in order to study centriolar inheritance in humans, for example.

You can read the rest, but there are clearly some very fundamental differences between rodents and humans

such that we cannot extrapolate directly from a rodent model to a human. I would say essentially the mouse is not an adequate model for human ART. It is a very good basic-science model, but you can't just go directly from a mouse to a human.

[Slide.]

I think that, because of these large differences, large physiological differences, in embryology, it is imperative, if we are to make progress, that embryo research relevant to human ART be conducted with nonhuman-primate models. But this is not happening.

Again, let's remember that humans and monkeys are both primates and share many similarities in reproductive biology, endocrinology and development.

[Slide.]

The United States is very fortunate, or we are very fortunate, because we have the best nonhuman-primate-research infrastructure in the world, by supporting eight regional primate-research centers and spending well over $50 million a year just to maintain them; not to do research, just to maintain them.

Although these centers are well utilized for disease research, they are hardly used at all for reproductive and embryology studies related to human fertility and reproductive problems. The reason for this is that federal research support for nonhuman-primate embryo research is almost nonexistent because NIH study sections refuse to recognize the value of these animals for studies on human reproductive problems.

Instead, these review panels tend, for the last two decades, to divert enormous amounts of support to mouse research, much of which is intrinsically good, but, at the time, they don't devote enough money, if at all any money, to nonhuman-primate research.

I would say that, during the past two decades, it is difficult to find any research studies relevant to human ART involving nonhuman primates or to find any mouse-based studies that have directly helped the practice of ART in humans. Again, I stress, mouse and other rodent research has provided a lot of very good basic data. The question is, can you apply that information directly to human ART. I think the answer is no.

As a result, the practice human assisted reproductive technology operates in a vacuum of basic knowledge about primate embryology so the success rates have hardly improved at all from year to year, only 1.4 percent from 1997 to 1998.

In addition, as you know, there have been some egregious lapses of judgment by some fertility clinics in applying novel technologies to human ART that have been inadequately tested in suitable animal models, if tested at all.

[Slide.]

Maybe I am just blowing hot air. So, to support this critique, I offer a sample of reviews on nonhuman-primate ART research proposals from two different NIH study sections. The first one states that, "Murine models are preferable because they reproduce rapidly, they are less expensive while there are few uses for nonhuman-primate-research models."

I think this reviewer has obviously summarized the situation very, very well and explains why reviewers favor the murine models and are against nonhuman primates. But this attitude directly contradicts the slide I showed

earlier explaining that rodents are not faithful models for human reproductive biology.

[Slide.]

This is a different review. This is of a proposal aimed to examine a fundamental property of primate embryos using monkeys as a model for humans, and that was to investigate intracellular pH which I think is going to be a major factor in embryo development. This is directly related to human ART. This proposal even proposed to use human embryo culture media.

Summary of the review; "The major weakness is the lack of important significance, either clinically or biologically."

So I want to emphasize that these what I consider prejudiced and hostile reviews of nonhuman-primate embryo research are the products of the NIH Center for Scientific Review which, in my opinion, has done an abysmal job of supporting research into human ART and infertility problems and which, thus, has done a grave disservice to the 15 percent of the U.S. population with fertility problems.

These kinds of reviews are not the responsibility of NICHD which has tried, for years, to support ART-related

research through targeted RFAs and other mechanisms. But, because the CSR reviewers dictate to the NICHD which proposals must be funded through the standard RO1 mechanism, we have a case of the tail wagging the dog.

Few, if any, CSR reviewers are scientists who work with nonhuman-primate models. In view of the strong reviewer prejudice against nonhuman-primate-embryology models, it is not surprising that very few proposals involving these important animals are funded by NIH.

Thus, we gather more and more information every year about mouse reproduction much of which is irrelevant to the human reproductive condition and learn almost nothing about primate embryology which is most definitely relevant.

As a direct result, a huge disconnection has been generated in the United States between the collection of basic data on reproduction and the clinical practice of human ART. I limit those remarks to embryology.

[Slide.]

There are several areas of ART or embryology most urgently in need of research to assist clinical ART, some of which are listed here. You can read these. I would add

to this--I forgot one that appeals to me, personally. I am trying to objective--but that is the measurement of intracellular ion homeostasis, calcium and pH which I am sure is going to become very, very important.

Two of the approaches shown here have already been applied to human embryos in ART without any basic data for guidance or indications for their safety, essentially performing experiments on human embryos that were replaced into patients without any knowledge of the likely outcome.

[Slide.]

My strong recommendations are for the federal government to heavily support nonhuman-primate research so that basic information on primate embryology can be obtained and new technologies for ART can be devised and their safety and efficacy tested.

I also recommend, if I had the power, to allow the NICHD to select the composition of the review panels that are telling them what to find. This is not novel. The USDA and NSF already do that.

I also suggest that the U.S. government support interactions between basic scientists and clinical reproductive communities to bring them together to close

the communication chasm that exists in the United States between these two groups. This latter action should take the form of supporting more--there are some already--symposia and workshops for exchange of information and technology which are present is occurring mostly through the generosity of drug companies with interest in human assisted reproductive technology.

Thank you.

DR. GIUDICE: Thank you for those thought-provoking words.

I would also like to invite Dr. Huszar to come to the podium. If you would share your comments with us and please limit it to five minutes so we have time for further discussion. Thank you.

DR. HUSZAR: While you get the slides activated, I would just like to remind you that we are all funded by the NIH and, actually, my research work on the spermatology which I will present, which I will show you, that, in fact the part of the sperm selection for ICSI, which is related to avoid mature sperm with aneuploidies, we have made a major success to make advances in it.

So, at least, in the ICSI, acception of the fact which actually don't buy the ICSI procedure, at least the sperm selection seems to be taken care of.

[Slide.]

I would like to thank the NIH, Phyllis Lepperd and also I see Michael McClure and also Donna Vogel, who is not here, who helped me during those periods while I had about five or six NIH grants subsequently after each other to support my research.

[Slide.]

What happened was that I came into the sperm and embryology from the outside of other areas of research. When I started to run the sperm lab in 1986, I realized that we should find, perhaps, something objective, a biochemical marker which tells us which sperm is fertile or which man is fertile.

What I have noticed, when we looked at creatinine kinase immunostaining, was that some of the spermatozoa were perfectly clear-headed and the other one had a different number and different amounts of creatine kinase which also was related to morphology.

[Slide.]

Essentially what happened was that when we looked in the a human hemizone, and we looked at the immunuostaining, we have found that, indeed, only those spermatozoa were binding to the zone which were clear-headed. That gave me the idea, which we proved the biochemical methods in a blinded study, that, indeed, the spermatozoa which underwent normal maturation and did not have cytoplasmic retention probably has a different type of membrane which is part of the sperm development process.

[Slide.]

Additional information came out of the data which has indicated that it was a protein, which we didn't know what it was, that, actually, we isolated that was an HspA2 chaperon protein which is known to be present in--you can see my immunostaining studies here--that in the presence of the spermatocytes where the meiosis happens and also during the cytoplasmic extrusion during the last phase of spermiogenesis are actually also expressed.

So there was a protein which gave an indication that there is a relationship between the meiotic process and also the membrane of the sperm to the extent that, if this is missing and the membrane is not formed and there is

no cytoplasmic extrusion, there probably are also some problems related to the meiosis.

In the paper of Biology of Reproduction, here are the summary results which kind of indicates that when you have an elongated spermatid, you have two ways to go. If you have the HspA2 present, then there is going to be cytoplasmic extrusion and the membrane is remodeled forming the zona binding sites and also a site for hyaluronic acid which we discovered which was very important because it coexpresses in normal sperm. These are the spermatozoa which have the normal meiotic process and have high DNA integrity which has been shown by a different paper.

But the important issue is here, that these spermatozoa have never fertilized before the ICSI events, the inception of ICSI, because of the fact that they don't have the zona binding site and they don't have the hyaluronic-acid binding site.

So my idea was, at this point, that we are going to make solid-state hyaluronic acid and see whether we can actually separate these two kinds of spermatozoa and actually select the spermatozoa which is normally developed.

[Slide.]

So here is actually--this is a plate which actually has been developed and created by a company, Biocote, who actually took the pattern from Yale and licensed it. You can see that the spermatozoa are swimming and it is coated in the area of the hyaluronic acid. These are the spermatozoa which presumably are the normally developed, and I will show you the data.

[Slide.]

In fact, here is the method that you can take that ICSI pipette and you can take any of these spermatozoa if you wish.

[Slide.]

Then I will show how this works. This is just an example of fluorescent in situ hybridization. This would be a normal sperm, that it has an X and Y sperm. Here is a disomic where you have one or two of the other and here is a diploid sperm where you have actually two components of the genetic material of the chromosomes.

[Slide.]

So here are the data. First, we have looked at oligospermic men. These are twelve moderate oligospermic

men which had a sperm concentration low but they were not ICSI patients because, in ICSI patients, you may not have enough sperm to have statistical significance.

Essentially what happened was when we looked at 50,000 sperm or so in the control semen and about 10,000 sperm which were selected by HA, that we had about a five-times reduction of the disomy four times, and the most important point here is that we had a 4.5 times reduction of the six disomies which, in fact, is an increase 4.5 times in the ICSI babies.

If you looked at the diploidy, we had about six to seven times reduction in the HA-selected sperm. This was very exciting. Then comes the next one which was even more exciting, that we do the same thing in normospermic men. The idea is that the embryologist looks at a very cute sperm and picks it up, and the question is, can you do it.

So here we have taken normospermic men who had an average sperm concentration of 120 million and we put through the sperm on the isolate gradient centrifugation which is the state of the art for sperm selection. When we have done, again, the semen spread and we did the HA-

selected sperm, we have found that the disomies, for instance the autosomal disomies, were reduced to halfway. The sex disomies still were reduced about four times and the diploid is reduced about five times, so indicating that, indeed, the HA selection works very well and that, indeed, that can be actually introduced in the clinical practice.

Also, this HA-coated slide can be used for reproductive assessment of male infertility patients because of the fact that, obviously, the sperm concentration does not tell you whether the man is fertile or the man has developed mature sperm.

Thank you very much.

DR. GIUDICE: Thank you very much.

I would like to thank all the speakers from this morning and invite our scheduled speakers to please come to the tables here to participate in discussion. So, Dr. Schatten, if you would join us up front, please, also.

Open Discussion

DR. GIUDICE: Clearly, from this morning, we have seen an overview of the challenges of assisted reproductive technologies including animal models, culture conditions

and I am glad that there was a mention about the ethics of culture conditions, and also the challenges of choosing the right embryo, choosing the right sperm.

Mention was also made about embryo implantation potential and early pregnancy markers as markers of poor pregnancy outcome. Preterm delivery and preterm labor comprise one area but there certainly are others because we know that women from ART cycles have increased risk of preeclampsia, gestational diabetes and so forth.

The safety and efficacy of the techniques are certainly very much in our mind and, also, the genetics associated with infertility, with regard to fertility, per se and also, on a larger scale, from a health perspective. So we have really covered quite a gamut of issues.

I would like to open the floor to questions. Dr. Schatten has a comment to make first.

DR. SCHATTEN: I would like to ask Dr. Racowsky and the audience a question and that is, in the ART community, we tend to do national or international experiments before we have any reasons or even evaluation mechanisms on these experiments.

We are conducting one right now in terms of aneuploidy screening. You spoke about concerns about whether there are any benefits. We hear from others that it appears as if there might be benefits. This is one example where I think, as a community, we should be able to step up to the plate and say, okay, what do we need to know now to advise our patients the best.

Are there mechanisms for the federal government to fund studies not on the actual biopsy of the embryo or the actual FISH, but the analysis of that data. Are there ways, as a community, that we can say, this is an important question. Lots of people have concerns. How can we go forward? And, if we can't go forward, well, maybe we should just enjoy lunch.

DR. RACOWSKY: I think you hit the nail on the head. What clearly we need is an organized group that is willing to coordinate the collection of data from multicenters. It is only if one does that and you get the numbers high enough so that you can really believe in them, that you can then start making reasonable decisions as to whether the test is appropriate or not.

We really don't know what the relationship, for example, is between Day-3 FSH levels and the incidence of aneuploidy. There are some papers that address it but they are not extensive papers. This would give us some idea, as a marker, for predicting the quality of the oocytes coming from the patient.

Perhaps there are subpopulations of patients who would benefit from aneuploidy screening but, as I said at the podium, I argue that, to take aneuploidy screening as a carte blanche test for all patients over a certain age who are undergoing ART is simply not appropriate and, in my view, not ethical because you are going to reduce the pregnancy rates, or the chance of pregnancy, of a large proportion of those patients because of the damage to the embryo, whether it be just a physical removal of a blastomere or, as we said earlier, with respect to the potency of the blastomeres and removal of blastomeres which will compromise the actual development of the embryo, per se.

So I would propose--and anyone in the audience needs to help me here, but I would propose a coordinated effort to bring together data from all centers in order to

address very simple questions such as the one I propose; what is the relationship between Day-3 and Day-10 FSH rates and aneuploidy rates in patients of different ages.

We don't know that. Until we have that sort of information, we are simply not in a position to really start evaluating whether the aneuploidy screening is appropriate for subsets of patients or not.

DR. BRENNER: Dr. Carol Brenner, University of New Orleans. I would also like to comment on the fact that we know if we have tested all chromosomes, there is probably a 70 percent chromosomal error rate in these oocytes and embryos. So here we are actually offering patients testing before we have even done all the experiments.

Again, I propose that we don't even do that until we actually assess all chromosomes and look at aneuploidy rate versus implantation rate.

DR. RACOWSKY: Then I think another point to be made, if I may, is the issue of mosaicism and whether aneuploidy screening down the road is actually going to be help us to be able to predict a developmentally competent embryo based upon the mosaicism that ensues following that.

DR. BAVISTER: Barry Bavister, University of New Orleans, also of Dr. Racowsky. Catherine, you gave a very nice talk and you told us something we already knew, but you explained it very simply and very elegantly and that is, when you stimulate a patient, you recruit a lot of follicles that are probably defective and were designed to become atretic and die off.

It seems to me that maybe all of the problems that one encounters in embryology with defective embryos and fragmentation and how do you select the best embryos and so on may be created by the fact that you are stimulating the patients to produce a very large number of oocytes, perhaps twenty, perhaps fifteen of which are doomed anyway.

Perhaps the approach needs to be to design stimulation protocols that produce fewer higher-quality oocytes. I don't think that is being done right now. That might make the embryologist's job much better and the overall pregnancy rate go up.

DR. GIUDICE: Dr. Scott?

DR. SCOTT: I think this can be highlighted, actually, answering Dr. Bavister's question, there are

three papers in the literature at the moment. I find them very, very depressing. It is taking this initial spurt of the whole blastocyst-culture concept to the next level where there are groups out there who have converted their entire programs over to blastocyst culture.

If you go and you actually analyze the data that is presented in these papers, it is very, very poor. There is about a 50 percent attrition rate of embryos growing out to the blastocyst stage and the maximum implantation rate of all those blastocysts--and this is now in nonselected groups of patients--is 40 percent in the younger patients. As you get into the older patients, it is lower.

That is in two groups. There is a third paper out there looking at natural cycles where the groups took natural-cycle embryos and they put them back either on Day 2 or Day 5. There was a 50 percent attrition rate out to Day 5 and the implantation rates of those natural-cycle Day-5 blastocysts was about 40 percent. On Day 3, it was about 20 percent.

On a per-oocyte basis, there was no difference between Day 3 and Day 5 and it was in the region of about 10 to 12 percent implantation which makes me go back to the

ovary and say that it is in the ovary. We have got to get more markers in our oocytes, understand how to look at oocytes and understand how to stimulate our patients and pick out the right oocyte because the bottom line is we still, even with the blastocyst, have an overall implantation rate of 40 percent which translates into a 60 percent attrition rate.

You know, there is so much work that needs to be done in markers there. That is just by dissecting out the data. That is where multicenter studies could actually help us.

DR. GIUDICE: Thank you.

Question in the back?

MS. PEARSON: Cindy Pearson, National Women's Health Network, for Dr. Schatten. You mentioned the data or the hints of data that you have in monkeys and possible behavioral effects. I wonder if you have done any statistics to calculate how many monkeys would you need to find actually significance in the differences and are there that many monkeys in programs in this country that could be looked at altogether?

DR. SCHATTEN: I think that is an excellent question. Ironically, we have the research resources in our national primate centers but we haven't been conducting the research. For example, Barry's monkey, Petri, is eighteen years old now. There are other monkeys that have been made by Don Wolfe's programs and other programs around the country. These represent an invaluable research resource that should be followed in a more comprehensive way.

To get at your question specifically, working with our colleague, Jim Sakat, at the Washington National Primate Center and the Mental Retardation Center there, he has done a power analysis and calculates that we need eight newborns of each sex; so that is eight females, eight males.

In the first analysis, I think what might be proposed is the comparison of natural or artificial insemination with ICSI and with ELSI, the ICSI being ejaculated, the ELSI being a testicular biopsy, and possibly also with embryos that have had blastomeres removed, as if in PGD or for PGD.

The reason I mention this, and this is so difficult because the work really is anecdotal at this point. When we did those first studies with five embryo splits, and here what we did is we dissociated embryos trying to make artificial twins.

We ended up with five offspring. Two of these offspring are dying right now from failure-to-thrive syndromes. One has Cushing's. The other has something we just don't know what it is. These are very small numbers. Maybe there is something about taking a newborn on I-5 from Portland to Seattle through rush-hour traffic. I don't know. But it is hard to know where to go with this kind of nonhuman-primate outcome data.

Is this something that should concern us or is this just, you know, bad karma. So, in the spirit of what Barry was proposing, the way we look at it is that you probably need around 64 offspring per year for a finite number of years, five years or so, and you need to follow them intensively two years after birth and then follow them as they go through life and aging, not with the same degree of intensity but just to follow them so that you know if there are any inadvertent outcomes.

We have a funny system with our national primate centers that, unless someone is paying the per diem, those animals tend to go into other studies and are lost. In fact, the taxpayer has paid a fortune for the gestation of those animals and it would actually be a modest expense to follow them later in life.

Thank you.

DR. GIUDICE: Gerry, I have a question and perhaps Barry could answer this or you can answer it or someone else in the audience. The point has been made that the mouse is not a perfect model for primates in terms of reproductive processes. I am wondering about behavioral.

DR. SCHATTEN: I wonder if I can first take that and then Barry can also address it. I think all of us support the enormous work that is going on on the mouse and nothing that I say, and nothing that Barry says, is to undermine the support for the mouse.

But I think, with mice and with nonhuman primates, you have what you need for the preclinical information. So there are tests of mice. For example, there is the water-maze test that Waterson worked out where mice learn where there is a little raft or they drown.

That seems Darwinian, but, anyway. Then you can test how long they remember that.

It is a reasonable test of cognition in mice. It turns out that mice that are produced from different culture media or also from ICSI, and this is unpublished work from Richard Schultz and Maurizi Bartolome, have challenges on the water-maze test.

Also, ICSI male mice of a very select inbred strain are less anxious than IVF male mice. Now, for my own family, I think I would prefer less anxious males but, as a population, we may not want to go there. It is challenging to know to extrapolate from a reduction in anxiety in an ICSI mouse to our own kids.

It is weird because I think all of us neurotic with our own kids. They are all mutations. Sorry. But to have clinicians either saying this work is immediately important for ART practice or rejecting it, I think, either underemphasizes or overemphasizes it.

I think, as a community of kindred spirits, we need to get the science, evaluate it in the context of what we want to know and then digest it ourselves before we left peer-review journals like USA Today cover it.

DR. GIUDICE: Thank you.

Yes; comment in the back?

MR. POLLARD: I am Colin Pollard, Center for Devices and Radiological Health, FDA. This is really directed to Dr. Biggers. I was intrigued by your comment that you thought it was unethical for doctors to treat embryos without knowing the specific concentrations. Actually, you were talking about specific concentrations of the individual components of media products from the various manufacturers.

That would imply to somebody like me, from FDA, that that is something that we should require manufacturers to do. It is not something that any of the professional societies have approached us about, ASRM, SART, whatever. Any particular thoughts?

The one thing that does come to my mind is there are academic interests that are different from clinical interests. That is certainly something that we at FDA have to consider as well as manufacturer sort of proprietary interest in these different products.

DR. BIGGERS: I don't think I can any more than what I said before. To me, I find it abhorrent using

solutions of unknown composition to expose embryos and cells.

DR. BAVISTER: May I add something to that? I find it also abhorrent, taking Dr. Biggers' comments one step further, that manufacturers may sometimes add components that they do not report, such as growth factors. Given the emerging information on gene expression in embryos being altered by different culture-media components, I think this is terribly alarming and I think it would be very much a concern to the FDA as well as to clinical communities to be using media whose composition they do not know.

DR. GIUDICE: There was a comment here?

DR. LEESE: Henry Leese, York, U.K. I will be talking about some of these issues tomorrow morning, but just one, as you move to follow up and also reducing the number of embryos for transfer, I would urge you to talk to the Scandinavians because they tend to be way ahead of the game here, the Nordic countries, Norway, Finland, Sweden, Denmark.

They are moving quite rapidly to single embryo transfer now, both in the fresh cycle and then in frozen

cycles. It may be significant that much of treatment is free there and so it doesn't cost the patient. Secondly, they are very strong on follow up.

There is a culture there that is different from here and in the U.K. that follow up is important. I think the sentiments about follow up is so laudable, but it will require resources. They will also require compliance. You could have, as it were, voluntary follow up.

In the U.K., with aneuploidy screening, it is mandatory because you get a license in the HFEA and you have report your results. So we can follow that up. The numbers will be very small and to do it on any big numbers, you need bodies like the IH, the ASHRA, the European groups, the Australasian. They are not research organizations, themselves, and so money would have to be found to do these studies--not vast sums, but money would have to be found.

DR. RACOWSKY: May I just make a comment regarding the single-embryo transfer. I think you have to put it in the right context. As a government and as a country, you have to agree what is an acceptable pregnancy rate per cycle. In Germany, the 2001 data shows that

ongoing--well, the actual live-birth rate from one ART cycle is 21.7 percent which is of the order of 15, 16 percent lower than our data I think will show in 2000.

DR. LEESE: It depends on how you express the data.

DR. RACOWSKY: This is by cycle start, 21.7 percent by cycle start. As I said earlier, their triplet and above rate is only 2.3 percent or so.

DR. LEESE: Yes.

DR. RACOWSKY: If the live-birth rate is acceptable for that country, then, yes, as a group, we can agree to go to single-embryo transfers now. But, given the constraints on our abilities to select embryos appropriately and the constraints that are defined by the still suboptimal culture environments, we are not going to be able to achieve the pregnancy rates that we achieve today in the United States with putting more embryos back if we do a single-embryo transfer.

DR. LEESE: I think is it compounded by the effects of multiple births, obviously. I think Anne McLaren had a remark on maybe European things. Sorry to take the chair for that one.

DR. GIUDICE: We have time, actually, just for a couple of more comments. Dr. Schatten and then one more in the back.

DR. SCHATTEN: Just a quick comment on this. All of our goal is one healthy baby from one egg from one embryo. There are different ways that we are trying to get from A to C and sometimes we are going through the rest of the alphabet. I almost wonder whether there might be a way to do a single embryo transfer on Day 3 and, within the next 48 hours, develop a marker to know whether that single embryo is showing any signs so that you could then have an option of either a second embryo on Day 5 or not.

DR. GIUDICE: That is part of the Holy Grail.

Yes; in the back?

DR. BUCK: Germaine Buck, Epidemiology Branch of NICHD. I would like to thank all of the presenters for a very thoughtful and exciting morning. I want to address the overarching concern that I think all the presenters have raised and that is the potential health and well-being of children and couples involved in treatment.

My perspective is a little bit different. I would like to suggest to the ART community that the

problems that you face with respect to following couples and children long-term really aren't all that different from other aspects of clinical care.

It is important to keep in mind that some of these issues about variations in practices, missing data, actually can be addressed with some of the newer methodological and statistical approaches that are available.

So I guess my comment really is twofold; one is that, to answer these questions, and I concur that these questions are paramount for our society, there will have to be collaboration across practitioners and sites. We have to get rid of this fear of ranking, even though it is done for other clinical treatment modalities, and also I think we need to begin to think about the minimal dataset that we really need to begin capturing and recognize that will change over time so that we can really provide the important information and give a full informed consent to couples engaged in these treatments.

DR. GIUDICE: Thank you for that comment.

The last comment by Dr. Lamb.

DR. LAMB: Can I just respond to that quickly. I hope that, in my talk--I was asked to address some of these problems but I really think that one of the major problems is to know what you need to follow up with. The reason we can't do that properly is because we don't know the causes of the infertility.

So my concern is that if we are looking simply as these babies for a year or two years, five years developmental defects, something like that, that is something that is fairly easy to measure.

But if we are looking at cancer development at a very early age, if we are looking at triplet repeat diseases, things that occur in midlife, it is very difficult to follow that up.

Now, those are two examples where we have good reason to be concerned based on other data that I didn't show you, but who knows what else to look at because, if you saw all of those genes, those were the tip of the iceberg. There are many, many people who have similar phenotypes but different gene defects leading to those same phenotypes.

So we don't have the technology to properly diagnose them right now. Therefore, we don't know what to follow up. I guess that was my bigger concern. The other things that I mentioned are problematic. They are not insolvable.

I really see the lack of our understanding of the infertility as being the major problem to follow up.

DR. GIUDICE: Thank you.

I think the comments, though, on the epidemiology and the techniques that are available are very, very relevant as on the clinical side, we tend to think that the waters are just too muddy to even deal with.

So I would like to conclude by thanking all of our speakers for a wonderful session this morning. We are going to reconvene at 2 o'clock. Thank you all.

[Whereupon, at 1:00 p.m., the proceedings were recessed to be resumed at 2:00 p.m.]

A F T E R N O O N P R O C E E D I N G S

[2:00 p.m.]

Social, Ethical and Policy Issues and ART

DR. WOOD: Hi. I'm Susan Wood, Director of the Food and Drug Administration Office of Women's Health. I am going to be comoderating this session. But, given the number of speakers and the way it is organized, my comoderator, Kristina Borror, will be moderating the first half and I will do the second half.

Greg Koski was not able to be here due to travel commitments that have come up but we are delighted to have someone here from the Office of Human Research Protection.

DR. BORROR: It is my pleasure to be able to moderate the first part of this session. We are going to have some very great speakers the rest of the afternoon.

When I first found out about this conference and the subject matter and the kinds of talks that were going to be happening, a lot of issues were brought to my mind about things that could possibly be discussed. Those include the boundaries between research and innovative practice and the jurisdiction that the federal government

has to regulate a lot of this practice and lots and lots of issues surrounding informed consent.

In addition to some of those issues, the first three panelists, in particular, are going to discuss issues surrounding kinship in the offspring of assisted reproductive technologies; risk-benefit, a weighing that patients and research subjects need to take into account when they are making decisions about whether or not to undergo some of these practices; issues surrounding oocyte donors; and, also, issues of justice.

So, without and further ado, I would like to introduce Alta Charo who comes to us from the University of Wisconsin Law School.

DR. CHARO: Good afternoon. I think some of you may disappointed, others may be relieved, that there is no powerpoint or other audiovisual aids for this presentation.

My involvement in this area I think of as being an object lesson to why setting policy is going to be so challenging for the people in this room and for the people in the general public who have an interest. Back in the middle '80s, when I was working for the now defunct Congressional Office of Technology Assessment, I became

what I think is, to my knowledge, still the only person ever funded by Congress to tour California sperm banks.

I got to tell you, working for the government is so much fun.

This was in the context of our OTA report on infertility and reproductive technologies. Within that report, although I was the legal analyst for the whole report, I was the study director just for the artificial insemination study that we did.

In the course of that study, I had occasion to come over to FDA and visit the infamous building up in Rockville, the Parklawn Building, to talk with somebody, and I really wish I could remember who it was with whom I spoke about FDA's plans back in 1987 for the regulation of sperm donation, just in case you think these things move quickly.

The conversation was fascinating because, as we discussed this, we realized that even with artificial insemination by husband, there were issues raised about whether to treat things differently when they were in a medical context as opposed to a purely private context; example, screening for viral transmission and potential

screening out from the suitable pool of people who would be helped with artificial insemination by husband those men who carried a transmissible virus. HIV was, of course, on everybody's mind at the time, keeping in mind that in a purely private setting, there would be no such screening or warning for the spouse.

When we move on to artificial insemination by donor, it became clear that it quickly got even worse because, while viral screening might be somewhat uncontroversial, as soon as you got into genetic screening of the donors, it got very sticky. Maybe everybody in the United States would agree that there are certain genetic disorders that are so devastating and so life-shortening for the newborn that it is simply pointless to have such a man be a sperm donor.

But there are a world of conditions that lie between that and what would be considered a benign set of genetic traits associated with a typical donor. As soon as the FDA considers anything having to do with genetic screening as opposed to viral screening, it is, per force, entering into government-based judgments about genetic fitness for parenthood, albeit, in this particular context

of medically assisted parenthood but, nonetheless, government judgment about fitness to parenthood. This was totally reactive.

It didn't surprise me that regulations didn't emerge quickly and it didn't surprise me when I saw the proposed rule on donor suitability that its focus was on things like viral transmission because this is, obviously, a difficult topic.

Because of that first experience, it doesn't surprise me, as we look across what I am going to outline as being several kinds of thematic areas of concern, that it is not an easy area for the federal government to intervene. I would also like to simply note that it is an area in which there are many other avenues for intervention so that not only federal intervention exists as a way to shape the experience of reproductive technology deployment in the United States.

I also want to say, by way of introduction, that I am going to outline asserted problems without offering my own judgment about whether these are real problems or not. I am going to leave that to the people who are focussed on the empirical information and want to make arguments from

their own sets of values about whether these are things we should worry about. I am simply going to note that these are areas that some people are worried about and outline some of the possible ways they can be tackled.

Let me take the first. Thematically, and it is particularly brought out by this morning's materials, there have been concerns raised about the speed of innovation in this area and the rapidity of its deployment in the clinical setting and the relative absence, it is asserted, of basic animal research or of carefully scaled-up human research supporting this.

Again, I am not going to make a judgment whether I think that criticism is valid. I just want to note that it is one that we have heard many times and will probably hear again.

There are a number of ways that this can be addressed, each one of which has its own special challenges. Obviously, one of the most efficient ways of addressing this is to have federal funding for the basic research because this provides an incentive for members of the academic and clinical community to begin to think in terms of rigorous systematic protocols that allow them,

while offering innovative care, to do it in the context of formal rigorous research that lets them maximize what they can use for the next generation of patients.

It allows for a more systematic approach and, in its idealized fashion, it may allow for each particular protocol to anticipate other protocols and collect additional information; for example, the presentations about genetic characteristics of the fathers in these infertile couples and the frequency with which there are various disorders, not necessarily being picked up every place around the country when they do their clinical care, is the kind of thing that might, in an organized research community, be routinely included as part of research protocols even where that is not really the focus of the research as a way to continue to amass rather comprehensive data.

This is, however, probably a foolish thing to be talking about because we know that, in the current political climate, there is literally zero chance of federal funding for this kind of basic embryo research when we are talking about human materials.

Now, the second possibility, and it was alluded to by Jill Warner this morning, has to do with direct regulation of these technologies via CBER. There are many things that CBER can certainly do. Of course, some of it is work on basic tissue practices and on facilities and even on facilities licensing. All very general good-practice materials are fairly unproblematic.

In, however, looking at a particular technology and asking is this a technology that has met the FDA's kinds of standards for a positive benefit-to-risk ratio, I think we begin to see some of the very special dilemmas that are posed in the area of reproductive medicine.

FDA does this all the time. When deciding when a new drug or new device is going to be approved for market, this kind of risk-benefit balance is typical. But it usually takes place in a very narrow environment in which the risks and the benefits flow to the same person. Although it does challenge our kind of libertarian myth about the United States, that the government has any role in preventing people from making a choice to take something for their own benefit even though there is very little chance it will succeed and a very high chance it will have

some side effects, even though it raises those, in general, we have come to accept the idea that the government has an appropriate role in protecting us from our own ignorance and foolishness in this very complex fear.

And it acts as a gatekeeper. Even there, not at all uncontroversial, looking at the drug recently having to do with irritable bowel syndrome; right? But, nonetheless, a very classic kind of balancing where the risks and benefits are to the same individual.

The dilemma posed by reproductive technologies is that the benefits flow to the prospective parents who are trying to conceive. But the risks flow not only to them in the form of failure or side effects, but also to potential offspring who might find themselves born with conditions that are unfavorably for a full and happy life.

How much of a risk, of course, is a problem because we can't answer it due largely to item No. 1 which is the lack of funding. We can answer it slowly more and more frequently now as we get experience from the clinical world and retrospective look at the data, but, nonetheless, there is this problem of a kind of asymmetric distribution of risks and benefits.

What to do. It does seem to me that this is an area where we might want to think about looking at other situations, and there aren't that many that came to mind as I was preparing this talk, but other situations where the risks and benefits don't flow to the same person and ask, how do we approach these problems.

There are two that came to mind immediately for me. One is very current and that is the vaccine situation in which one of the dilemmas about the small-pox vaccine is that, although it confers a benefit of resistance to the person who receives it, not only does the recipient have a side-effect risk but there are people around the recipient, particularly immunocompromised people, who now are going to be at risk by virtue of somebody in their vicinity having just been vaccinated, and how do we balance this kind of risk and this kind of benefit.

Or, perhaps, considering that so many of the benefits of helping people to have children with these techniques, helping people to have children gestationally or genetically or psychological and emotional, maybe it makes sense to look in the psychiatric area and look for examples of drugs in which there was a tradeoff between its

efficacy at making somebody no longer any kind of threat to society--this is, obviously, a very small, tiny portion of the mentally ill community--versus the benefits of the drug to the individuals, themselves.

Do we have examples in which we were thinking about public safety being traded off against the drug being more easily tolerated or somehow otherwise advantageous fort individual.

But it is this kind of dilemma that I think lies at the heart of the risk/benefit analysis here and because, in that risk/benefit analysis, the FDA would necessarily also have to make some value judgment about how important it is for people to have children that they are either gestationally or genetically related to, a value judgment that is really not one that is amenable to some kind of scientific inquiry but is really very personal judgment and one on which there is wide diversity of opinion in the United States.

I would hazard to guess that even what seems like the most classic of FDA activities which is to take something like, for example, cytoplasmic injection. I forget the terminology you are now using, ooplasmic

donation--to take just that one example and look at it and ask what would the classic kind of thing be; it would be to do a risk/benefit analysis and ask is this really ready for prime time.

Even that would not be all that simple because, buried within it, is this dilemma of the balancing of benefits to the prospective parents and how strongly do we value that benefit. That has been a fight within the infertility community for years, to get people to take their problems seriously and, at the same time, how do we, then, take whatever value we assign to the fulfillment of their aspirations, how do we balance that against risks that are imposed, in part, on third parties that have, in no way, been party to agreeing to this kind of endeavor.

Before I move on from this question of the introduction of techniques, let me just mention that there were two other, at least, areas where you can have a strong influence on the speed and style of introduction of techniques.

One has been hinted at here and that is postmarketing surveillance or some kind of post-care surveillance which is not forbidden under any kind of

restrictions about embryo research--here are certainly no restrictions on the use of federal money for this--and might help to clarify, for a number of people, whether or not there are significant problems occurring at an elevated rate among the offspring or among the parents who underwent these procedures and put to rest some of the questions in this area that continually get asked.

The last, and this is one where I know that everybody here with an M.D. is just going to go whew and they are not going to want to listen to anything else I say, is the tort system. For those people whose goal is to severely slow down the rate of innovation and deployment in this area, a very effective tool is to work with the tort system.

Currently, medical malpractice is handled under a negligence standard. That means that you ask did the professional act in a way that is consistent with the standard practices of the field. In the reproductive technologies area, that is a very difficult question to answer because the standards are constantly evolving. In some ways, you can almost say that there is almost no standard with the really frontier treatments like the first

cytoplasmic injection. How could you have a standard for something that is absolutely new?

So, using a negligence standard, coupled with the fact that the prospective parents typically, even given the imperfect information, will say, I accept the imperfect information and accept those risks, acting as a kind of block in the causal chain of events and, finally, outcomes which may not appear for many, many years, thereby running into some technical problems having to do with statutes of repose and statutes of limitations in some states, collectively means that the medical malpractice system based on the negligence standard does not really do much by way of regulating through the tort system in this area.

You should recognize that the tort system is about regulation. It is not about regulating by directive. It is about using the economic effects of litigation and judgments and settlements to create a new set of incentives and disincentives for behavior. So it is a form of regulation.

Imagine, if you really wanted to shut down innovation fast, what you would do is you would move from a negligence standard to a strict liability standard; that

is, we know that there are always bad outcomes despite the best possible practice in everything, whether it is in blasting a building or it is in driving your car.

But, when it is blasting a building, we use a strict liability standard in which, if there is a bad outcome despite every bit of care, you still wind up having to pay the damages. Why? Because there are some areas of social life and activity that, as a society, we have decided, through our courts or our legislatures, are things that ought to internalize all the costs of the injuries that they spread around the world because we only want them to proceed if they are so valuable to society that they are not only profitable, in and of themselves, they are profitable even when they take care of the people who were injured despite their best efforts. No moral judgment here; purely an economic judgment.

If you had strict liability in this area and if you coupled that with the removal of these kinds of obstacles having to do with the parental consent or the statutes of limitations, what you would have done is you would have said to practitioners, you are not going to do this unless you are so confident that you are going to have

a good outcome that you are willing to take the risk you will have to pay for a bad one.

That is a huge disincentive. In the end, because everybody is insured, it would mean the insurance companies become the regulators. So, again, without suggesting that there is or is not a problem in terms of the rate of innovation here, I would say that that, for people who think that innovation is out of control, needs to be stopped, we want to shut all these things down, doing the nth degree of tort reform is probably your most direct route.

For those that wish to simply increase the quality of the information and allow for the usual typical kind of medical self-regulation through professional societies and professionalism among members, it would be federal research monies, not soon, and postmarket surveillance.

For those that want to take a stab at having some government intervention in the speed of particular technological innovations, it would be through direct regulation of something like a particular new technique granting that there it will not be easy because there will

be tremendous debate over the value judgments that are embedded in those risk/benefit analyses.

Now, let me move on to some of the other problems--I see I have used about fifteen minutes of my time--because there are a number of them and some of them really don't have much at all to do with FDA in particular and sometimes not much to do with the federal government.

One of the more widespread concerns expressed in this area ever since the early 1980s is something that gets at a kind of mish-mash of exploitation, commodification of the human body, commercialization of human emotion and the distortion of family relations.

I put them all together because they are all kind of mutually reinforcing concerns but they could be separated if we have as much time as like a semester course of reproductive technologies.

Again, without suggesting that I agree or disagree with these concerns, let me just say they are going to be raised over and over again. Again, there are many ways to begin to address them. One is to look at, from the point of view of the tissues and the body, the way in which body tissue is moved from person to person.

We have a system now that calls itself donation in which some payments are made for actually out-of-pocket expenses. Other payments are made that are substantially larger and probably do serve as incentives that go far beyond the usual out-of-pocket. This is happening in the context of sperm where the production and collection is not painful--embarrassing, perhaps, but not painful--to eggs where it is very uncomfortable and where there are repeated questions being asked about the long-term health effects, although not yet confirmed that there are intermittent use of these drugs.

Finally, surrogacy in which we are talking about the most profound use of the body because it happens over many months and has, of course, a particularly significant emotional impact.

Now, if you were to regulate or prohibit the sale--let's just talk about tissues--of eggs and sperm, you would have a tremendous impact on several of these items having to do with commodification, et cetera, that I was talking about. But the question is what is the goal. Is the goal here to regularize the procedure, to facilitate the deployment of reproductive technologies while

simultaneously eliminating the actual or the possible problems of exploitation and distortion of human emotion, et cetera?

If it is simply to regularize while minimizing the possibility of these other problems, then, certainly, you might be looking at regulations that have to do with the scale of the payments or the kind of information people have before they are allowed to decide that they want to have their sperm or their eggs used by other people.

If your goal, however, is to severely contract the scale of reproductive technology activities in the United States, what you would want to do is you would want to ban all monetary transfers here. Doing that means that, although you will not shut it down and you will not be making it illegal, you will certainly make it much more difficult.

You will also do something else that is rather interesting because now, where people can obtain gametes from strangers that they never meet because of the system of donation-sale that we have, you can have people engaged in these activities with a minimum of emotional involvement

with these third parties who remain these kind of amorphous invisible creatures.

But if you moved away from payment for gametes, chances are you would move to a system in which most gamete donation would come from people who are known to the couple; family, close friends. That means that you add a whole new emotional dynamic. There are people who would argue that that is an excellent thing because what it does is it recreates the actual emotional dynamic of family making.

Just as when you have sex outside of marriage and you might conceive with somebody, or you conceive with somebody and then you divorce them, and life is messy and families are messy and these are the emotional ties that bind and that that is a good thing, other people would argue quite the opposite and say this is a horrible thing.

The same argument, by the way, goes on in the area of organ transplant in which some people have argued that organ transplant from family members is a horrible thing because of the emotional pressure that the healthy family member feels to step up to the plate and be altruistic and allow his or her body to be cut open and a

kidney removed or a liver lobe removed, et cetera, and that this is really a way to manufacture dissent in families where there hadn't been any.

So very different results. But, again, try to think through what your goal is--is it to shut down this area or is to simply regularize it will determine what you want to do in the area of the market in human tissue.

Similarly, with the contracts for surrogacy. If you want to regularize it, which certainly helps people, no matter what the rules are you pick--almost any rule is better than no rule because chaos is more emotionally and legally draining than certainty. If you want to, therefore, facilitate and regularize, you would regulate the contracts, make it very clear when they are enforceable, when they are not.

If what you want to do is slow this particular technique, what you are going to do is either leave it a total wild west or you will prohibit any kind of enforceability for these contracts.

Note, by the way, that whereas the sale of tissue is, in fact, a federal question although one that would have to be handled with legislation, not just by

regulation, things having to do with contracts and family law--and, by the way, defining the kinship relationships also falls into this category, whether your goal is to regularize or to deter. Those are state law questions. So a state that wants to shut down this industry a little bit, they are going to write their state laws and make it very clear that, no matter who carried somebody to term and no matter who intended to be the parent, that you can never, ever, ever, ever get the genetic parents out of the picture. You can never, ever, ever, get the gestational parent out of the picture. In other words, you can't use private arrangements to make certain people who were biologically involved in the conception magically disappear.

Other approaches, however, don't want to use kinship law to both break new ground in family law or to slow these technologies but, instead, simply want to regularize so that people can proceed with a minimum of fuss and a minimum of uncertainty which, by the way, is probably a good thing for the kids who are often at the center of the battles that develop.

Looking at the time, I am going to skip over the next which is the commodification of embryos and the destruction of embryos and the dispositional authority over them. I suspect some of the other speakers may touch on this but I am happy to come back to it afterwards.

Let me just go to the last kind of big area that I think is one that is not particularly appropriate or amenable to a quick fix, particularly an FDA fix. That is what I am going to call the kind of eugenics concerns. They really take on two different forms. One is the selecting against form; that is, people who are going through IVF and have extrauterine embryos now have an easy add-on. If I am conceiving through natural intercourse, it would take a lot to make me want to abandon what is fun and cheap and private and do something that is expensive, difficult and in a doctor's office.

But if you are already going through IVF and you have already got the embryo outside the body, here is an easy add-on; let's do PGD. So this is certainly going to be the first target market.

Question; PGD for conditions that are lethal or severely disabling early on in the newborn's life, there is

some distress in some quarters at the thought of any embryo not being used, but there is not a lot of distress about that particular choice once one accepts the idea that every embryo is going to live.

But, of course, with the announcement, oh, it must be eight months or so ago, about the use of PGD for the prospect of an early-onset Alzheimer's, approximately thirty, forty years down the road. This was all probablistic, of course--came a kind of reprise of the debate about what is going to happen if we go too far.

What is it that we are going to be screening against? Are we going to be screening against everything? Of course, I follow this with great interest knowing that if they could have diagnosed half the things I have got, I certainly would have been eligible for that kind of screening, myself and I suspect a fair number of you all as well might have been screened out.

The trouble is this is not really an FDA issue; right? You can check a technique like PGD for its ability to accurately diagnose a particular condition or to accurately diagnose it without reducing the viability of the embryo unduly. But once you get into the question of

appropriate uses, which you might be tempted to do in the risk-benefit area, you are now very squarely into an area that is really about social judgments as opposed to risk-benefit judgments.

We have a lot of history here. In the abortion area, still on the books in much of Continental Europe, is technically the notion that abortion is a criminal act that is excused "only if." In some places, still, "only if" certain kinds of particular conditions are met or certain kinds of sign-offs are obtained from doctors.

So we know that the same kind of thing took place in the United States. We know, in the area of artificial insemination by donor, that the physicians acted as gatekeepers to single women, to gay women and particularly to single gay women, noncoupled gay women, saying that this is really not for you.

Yet, these are judgments about the social aspect of this medicine; that is, who really has some kind of entitlement to be a parent. Is it anybody? Or is parenthood a privilege? I am not talking morally, I am talking now legally, because we can all agree that

parenthood is a privilege when it comes to talking ethics, but that really doesn't get us very far.

But is it a privilege or is it an entitlement? If it is an entitlement, then how we make the distinction between those for whom it is no longer an entitlement because they have a characteristic that we consider to be too dangerous for their children or because they are too unwilling to accept children however they may turn out without this kind of screening.

In the end, notice, because, in the selecting against, unless you are going to believe that it is a harm to any embryo not to be selected, and that is different-- but if you are going to look at harm as, like, living with a problem, the selecting against doesn't harm anybody directly. A child is born free of a particular condition because that is the way that child's embryo was constructed. The child who would have the condition just never gets born.

So is about harms not to the children, but it is harms to society. It is a harm to the collective morality of the tolerance for people with disabilities, et cetera.

Similarly, the flip side, the selecting for and, in the more kind of science-fiction notions, the enhancement.

Again, no harm to a particular child who is born after having been specifically selected or enhanced; the concern has to do with the exacerbation of inequities in society, the concern that we will have a class of people who have access to all this fancy medicine and are going to wind up producing only children who are not only free of defects but, at a future day, are going to have certain qualities enhanced and the rest of the masses will produce the plebeian underclass, a biological and well as social underclass.

But these are questions about social inequality that may not be possibly addressed at the level of federal regulation over the particular technologies. If they were, we would also be equally worried about a number of technologies that undoubtedly do give biological advantages to children of well-educated or upper-class parents, and that includes the use of nutritional supplements during pregnancy or even extended breast-feeding by women who can take more time off from work before they have to go back to confer enhanced immunity for the course of life.

Or even extra visual and auditory stimulation in newborns which seems to be associated with life-long gains in learning capacity, not merely short-term gains in learning capacity.

So we have a huge dilemma here because one of the things that worries people the most about these technologies is the very thing that you cannot address through any single intervention, whether it is tort law or FDA regulation or federal research. It is about a kind of social-climate issue. That is a much fuzzier issue that must be addressed in myriad ways having to do with Hollywood, having to do with schools, having to do with churches and synagogues, et cetera.

Let me conclude with one other very specific example because it highlights, I think, one of the dilemmas here and presages tomorrow morning's presentation on the HFEA which recently had to deal with this, and that is a very special kind of "selecting for" and that is selecting for an embryo because you think that the resulting child might be a potential donor of tissue to a sick sibling.

This is a case that has occasioned a lot of comment in the United States over the years as well. A lot

of people find this a very disturbing scenario and, indeed, are even moved to say, "You know, this is just why we need something like the HFEA in the United States, because we need to have some kind of collective judgment this is bad, and tell people that they can't do it," and have that enforced through government action.

I would hazard to guess that it is more complex than that because, buried within the judgment that this is a bad thing is a kind of Kantean notion about respect for persons; that is, a notion that it is wrong, it is disrespectful, to treat a person as a means rather than as an end in and of herself, which all sounds really good.

But you do need to keep in mind that this, too, is not ex cathedra. This is very much part of the post-Enlightenment Continental version of individualism and there are whole other worlds of culture and religion that view the worth of the individual not only in terms of the individual being treated as an end in and of herself or as being particularly entitled to have complete freedom of action, but also consider the worth of the individual and the nature of respect as somehow embedded in the ability of the individual to serve others that would see the ability

to be born and, at the same time, provide this life-giving service as akin to a "blessing" and not, somehow, an imposition.

So, again, just keeping in mind that that kind of attitude which, by the way, I find buried in my own tradition which happens to be Jewish rather than Christian, and I think I have found in some of the writings from people coming from Asia, makes me suspect that, as we recognize the diversity in the United States, religious and ethnic and national origin more and more thoroughly, we may also come to expect more than I think we do now, disagreement over things that seem perfectly obvious.

Another example, the playing-god notion, which makes perfect sense within certain kinds of Christian theology but makes no sense at all in Jewish and Islamic thinking where taking action is not playing god. Hands off, letting nature decide, would be playing god and taking action is playing human.

So, again, I don't believe that it is an easy thing to contemplate an exclusively federal response to the range of concerns that are raised in these technologies because embedded in that federal response will necessarily

be some very significant value judgments about the entitlement to be a parent, the degree of pain that should be respected in conjunction with the experience of infertility.

The degree to which there ought to be adult sacrifice of personal interest on behalf of embryos, fetuses and prospective children, the degree to which the change in family relations, the move from state-oriented declared family relations, be it marriage, et cetera, to privately order family arrangements through contract arrangements with surrogates and sperm donors and egg donors, whether or not we want to discourage these or encourage them, give them great weight or little weight, are all exceedingly controversial value judgements.

I don't sense, in the United States, enough of a consensus nor a constitutional culture that would let that consensus be imposed upon a dissenting minority such that it would be a trivial task to set up something like a regulatory authority. Targeted interventions on purely safety issues, themselves, I think raise enough questions to keep us busy for a good time to come.

Thank you very much.

DR. BORROR: The next speaker this afternoon on the social, ethical and policy issues in ART is Andrea Bonnicksen from Northern Illinois University.

DR. BONNICKSEN: I would like to thank very much the organizers of this meeting for inviting me to speak. Also, I would like to acknowledge the importance of their mission. Informed consent is a cornerstone of ethical medical practice and a core of expectations has developed about informed consent for medicine in general and for ARTs in particular.

Today, I will look at the matter of informed consent for a particular type of ARTs; that is, the innovative ARTs in their first uses in the clinical program.

[Slide.]

I have been asked to address several issues and I selected among them these three. First, I will look at protecting the interest of oocyte donors; second, making informed decisions when data are inconclusive; and third, making informed decisions when interventions may result in inheritable modifications.

I will use one innovative ART and that is ooplasm or cytoplasmic transfer to illustrate the things that we might keep in mind as we think about protecting the interests of parents and potential children if ooplasm transfer were to proceed in the clinical setting.

[Slide.]

Protecting the interests of oocyte donors; core expectations have developed about informed consent for oocyte donation. For example, the American Society for Reproductive Medicine has published guidelines for obtaining informed consent in ART in general and also in oocyte donation, in particular. Then it has developed certain guidelines for particular issues within oocyte donation such as the matter of payment.

The NABER, the National Advisory Board for Ethics and Reproduction, which is no longer in existence, RESOLVE, academics and others have also developed very thorough guidelines. There is a website, the University of Michigan Reproductive Sciences Program, for example, has a nice document on it, What Every Woman Should Know Who is Thinking about Being or Using an Egg Donor.

These are some of the topics that were covered in the 2002 oocyte donation guidelines of the ASRM such as indications for use, psychological evaluations, the screening and testing of donors, multiple donations. This illustrates that informed consent is part of a program's overall policies about what should be offered, when, and under what conditions.

[Slide.]

The guidelines to this point for oocyte donation have been developed with whole oocyte donation in mind; that is, the use of the entire oocyte where the donor's nuclear DNA forms the female's genetic contribution to procreation. With what I call whole oocyte donation, the donor is the genetic but not the social mother of the offspring.

Ooplasm donation is slightly different because it would use only a small part of the oocyte, approximately 10 percent of the ooplasm and not the nucleus. Ooplasm transfer or OT led to the first birth in 1997 and to approximately 30 births as of March, 2002. The aim was to circumvent problems of embryonic development.

It is now in effect on hold as the FDA has required an IND to be submitted for review prior to making a decision about the--before proceeding into the clinics. If clinical trials commence and if oocyte transfer is shown to be effective and safe, it will likely be an attractive prospect for women of advanced maternal age.

Cytoplasm from a younger donor could help rejuvenate the recipient ooplasm. A number of people, for example, have talked about egg rescue and this might be a nearly irresistible idea for some women of advanced maternal age. If this happens, then, there may well be a growing demand for oocyte donors.

[Slide.]

On the first day of classes in the fall every year at the University where I teach, the student newspaper has an ad, a large ad, in big bold print that says, "Egg donors wanted. Compensation, $5,000. Apply here." I bring this up often in the classes just to kind of point it out and the female students in there seem taken aback about the idea of donating because they are not sure they like the idea of having children who are genetically related to them that they are not raising.

What I have here is a highly simplified schema that compares the whole oocyte donation with ooplasm donation because I am suggesting here that whole donation brings with it relatively high stakes, because women who would want to help others conceive or who would like the idea of having genetic children out there would get a high psychic reward from being an oocyte donor. Yet, at the same time, they set themselves up for a potentially high emotional risk if they later fail to have children or if they begin to wonder if they had been genetically related to some children they don't know about.

On the other hand, those who are donating or who would donate partial oocytes for ooplasm transfer could experience altruistic rewards by helping others conceive but they will not have the same emotional risk because no genetically related children would be born.

The health effects of ovarian stimulation and retrieval would be the same for both groups presumably, I am not sure. It is possible that not a large number of eggs would be needed for OT. So all of this is to say if OT is offered and it works as expected, it might be an inviting alternative for prospective donors.

To handle this, the guidelines for donation that have been developed for whole oocyte donation would provide a starting point. For example, on the question of compensation for whole donation, the ASRM has recommended that payment should not be based on clinical outcome. That means that the same principle, one would presume, ought to apply with ooplasm transfer, too. If a pregnancy does not develop or if the embryos do not develop, the donor still will be compensated.

Another principle, or one of the core expectations, is that monetary compensation should reflect the time, inconvenience and physical and emotional risks and demands of the donor. Some might argue--I haven't heard this yet, but I would presume we might sometime hear it--that the amount should be less, the amount of compensation should be less for ooplasm transfer because the egg, without genetic material, is less valuable.

But it is still the risk to the donor, not the value of the oocyte that should be determinative. The donors who undergo the same procedures ought to be compensated in the same manner and this also would guard against the idea of eggs as valuable commodities.

Another question would relate to the payments. They should not be so large that they would be undue inducement.

[Slide.]

Another question about the potential limits. Should donors for ooplasm transfer be limited in the number of times they can donate? Whole oocyte donors, at present, are limited partly because of the fear or concern about inadvertent consanguinity. But here that would not apply. So then there is the question of whether the limit should still be enacted and would it be based upon health risks.

Another question; should ooplasm transfer donors be limited by age. The ASRM, at present, has recommended that donors be between the ages of 21 and 34 with recipients advised if the donor is older than 34. So the question could be revisited.

Previous motherhood; for whole oocyte donation, proven fertility is desirable. Should this apply for ooplasm transfer as well? These are all questions to be considered. Other issues; family pressures to donate might be the same for both groups and the clinics will need to screen and test for mitochondrial diseases, infectious

diseases, and others and it should be clear to the donor whether the information would be available to her afterwards.

Donors for OT should know that they will not be contributing their nuclear DNA, just in case they are confused and they want to have genetically related children. Conversely, donors who are thinking they are contributing their whole oocytes should not be used for OT unless they have specifically given consent for that.

So, in summary, the core guidelines are a starting point here and they can be modified to be responsive for issues arising from OT or other new uses of oocytes in innovative ARTs. Here, we can kind of guess what some of those uses might be.

[Slide.]

Another question relates to making informed decisions when data about safety and efficacy are inconclusive. Unknowns are a part of medicine. Uncertainty pervades all experimentation and innovation within medicine. Yet the inconclusive data for ARTs attracts special attention for several reasons. Among these are the health of the children being at issue, the

concern about patients being especially vulnerable because of their yearning to have children.

Patients often pay out of pocket which means that a technique that is inefficacious, if it is applied, then the patients are being penalized. There was concern about the lack of abundant animal data when ICSI was developed, as we heard earlier this morning. The history of not funding human embryo research; all of these things have created concerns about whether the techniques are being introduced too quickly.

[Slide.]

Some questions to ask for patients who are trying innovative technologies; will this pose risks to my child, to me. Especially with older women, using OT, for example, if that is used, for older women, then all of the risks of older pregnancy should be considered as well.

The question about whether the benefits have been documented, the harmful effects, if they have been documented, and, more specifically, will this benefit someone with my particular condition. As we saw this morning, many of these things--I mean, there are so many

unknowns that it is going to be very difficult to answer these questions specifically.

[Slide.]

More questions to ask; what is the clinic's experience with the procedure, what are alternatives to the procedure including adoption and non-treatment.

[Slide.]

Some of the things that could help with making decisions when data are inclusive; one would be the access to clear and manageable information. What is happening now with the governmental oversight is to try to set up the stage where more data will be gathered systematically ahead of time. That is only part of the equation. The other part is how to translate that to information that would be useful to patients and not so abstract that it would be very difficult for them to understand.

Also the challenge of the neutral information and interpreting animal-based studies. I don't know how that--it is something to be studied, to determine how people hear that, if they hear that mice models have been used or other animals, how do they translate that mentally from animals

to humans and what are the best ways of making this available to them.

Also understanding the status of the procedure, whether it is experimental in nature. The styles of decision-making involving risk. It would seem here that one could turn to the literature among genetics counselors to determine how patients hear risk and what they do with it afterwards.

For example, if one is told that one has a one-in-four chance of having a child with a serious disorder, that sounds much different from being told, you have a three-in-four chance that the child will not have that disorder.

The styles of decision making involving the interactive consent process. Here, coming back, rather than just giving the information, asking what is your understanding of this. That could help in the understanding of all parties in the process. And the question of deciding who pays.

It seems here that a model might be the Fertility Success Rate and Certification Act of 1992 which involved in a partnership among the Society for Assisted

Reproductive Technologies, RESOLVE, the CDC, FDA and other governmental agencies systemically to bring together information and to convey it, and the information that comes through that is available on the Internet and other places, is very clear and has the hand of a number of people involved in that. So this could be a model here.

[Slide.]

Making informed decisions when interventions may result in inheritable modifications. I distinguish here between two categories of inheritable modifications; the alterations to the nuclear DNA which, of course, has been performed with animals. It is not yet on the immediate horizon with humans. Then alterations to the cytoplasm which would involve mitochondrial DNA with at least two children who it has been determined that they have the mitochondria of the donor and the recipient in their cells differentially, at least.

[Slide.]

There are different perspectives on ooplasm transfer and I have labeled them three categories here. One, the permissive, where the idea is that it is possible now to proceed with existing oversight mechanisms. A

cautionary approach; proceeding may eventually be possible--that is, it is not precluded--but with heightened oversight. Third, that it should never proceed in a prohibitive category.

[Slide.]

Going back over the permissive. Here is the idea, and I am putting this together, that the inheritance of mitochondrial DNA is not automatically troublesome. As a matter of fact, in one of the articles written that reported the heteroplasmy of the two infants, the authors felt that this was a positive, not a negative, sign because, as they say, it demonstrated that the transferred mitochondria can be replicated and maintained in the offspring, therefore being a genetic modification without potentially altering mitochondrial function.

They foresee benefits that are rather broad for older women, women of advanced reproductive age, as well as for problems with infertility; that is, perhaps this could be a court of first resort rather than the court of last resort.

The question about the balancing that we have heard previously of the individual and society, that this

is primarily an issue of autonomy. The implication of this kind of approach is that consent may be given when safety and efficacy are demonstrated. I should have added here, when IRB approval has occurred.

[Slide.]

The cautionary approach suggests that the inheritance, per se, of mitochondrial DNA is troublesome and there are benefits; that is, one can visualize these but they would be more narrow, only following very select clinical criteria, and here OT should be a court of last resort rather than first resort.

The societal interests are important here and should be weighed against the individual interests. The implication of this approach is that consent eventually may be given if the conditions are met. There were two articles in Science, one by Eric Perins and Eric Youngst and also by Mark Frankel and Audrey Chapman that, in essence, took this kind of approach by suggesting that consent eventually may be given but one needed data collection first, a new oversight body or the IND process and also a public discussion.

[Slide.]

Then the prohibitive approach is that the inheritance crosses the line. It sets the stage for nuclear DNA alterations. It changes the mind set. It moves us into a field we should not go. The idea is that there is no clear benefit here because there are less problematic alternatives available.

When one weighs the societal and the individual interests, the societal interests prevail and the implication is that consent can never be given even if safety and efficacy were assured

[Slide.]

Making informed decisions. If OT were to proceed under the cautionary approach, and I would assume that this meeting and other things indicate that it would be a cautionary approach rather than a prohibitive approach. One could start off with the core informed-consent guidelines; that is, there is a core that already exists. Work off of that.

Looking at animal data across the generations, making the decisions of how many generations need to be tested and followed. Access to clear information about the data that is reported in the IND because an IND would bring

together a great amount of data. Again, how to translate that to something that is meaningful.

We heard this morning that practitioners might take a word and scientists might take a word and they interpret it differently. We could add patients to that, to say that patients would use a different word altogether.

The question about emotions; if a child's health is compromised. On the Internet, ivf.com, looking at a consent form, a sample consent form that was recommended for ICSI, one of the phrases was, "We understand that we may experience emotional distress, conflict or regret should the outcome of the IVF ICSI procedure be less than optimum and the child inherited problems that either were or were not predictable."

So the potential parents would be looking to how they would feel if a problem resulted and also to the reproductive capacity, let's say, of the child. So it seems to me that patients can proceed, or could proceed, with proper protections because patients who use ARTs are giving consent on behalf of their children and people who do not use ARTs and who have children who are at genetic risk also are giving consent on behalf of their children.

The picture gets complicated when one tries to say that some risk-taking is appropriate and other is not. The preferable approach is to make sure that this is as safe as possible and that couples are aware of the risks and that the discussion be broadly conducted.

[Slide.]

So, in conclusion, building on informed consent for innovative ARTs, overall the informed-consent system is in place and developing the best scientific processes for ARTs are now under way. That is a meritorious goal. It also deserves creating thinking to ensure that the implications of the evidence-based approach are understood and heard by patients.

The challenge, then, is to promote understanding when the understanding is elusive to physicians and clinicians and researchers as well as to patients.

DR. BORROR: The next speaker, and the last one before the break, is Mary Mahowald.

DR. MAHOWALD: Good afternoon. I hope I won't hold you past the break. It is good to be here.

[Slide.]

I thought I would like to begin with my favorite cartoon about reproduction. I often use this with medical students and I ask them to note that the process is not as easy as it looks and that one person really does most of the work.

[Slide.]

Then I ask them what the traditional recipe for baby-making is and they don't have too much trouble answering that question with these as the basic ingredients. Then I suggest that if and when most of them want to be parents, about one in ten are going to find that the usual recipe doesn't work.

[Slide.]

So they might pick up a cookbook that has some other recipes like these as a partial index of possibilities. My suggestion is that at least one criterion by which to judge whether one of these alternative recipes is a good one or not is to check how far it moves from the traditional recipe, how much it varies from that.

I think our task over these few days is kind of similar and complementary to the one that I pose to medical

students to assess current and anticipated recipes for having children and to suggest ways by which they should be facilitated or possibly restricted.

Like other speakers, I have some problems with terms. In the first part of this talk, I am going to identify some of those that I think are problematic without going into them in any depth. Then I am going to talk a little bit about what I consider, and I suspect most of you consider, the central principle, ethical principle, that ought to underlie any regulation that this group or any other group develops around assisted reproduction and that is principle of justice which, granted, is open to different interpretations but which, I will argue, demands of us attention to very important differences of cases.

I will offer some examples of differences that deserve attention and the research areas that I think deserve support. I will conclude with just a list of suggestions that can be considered components of recommendations or policies in this area.

[Slide.]

So here is my list. It is just a beginning list of terms that I think deserve analysis. The first one, in

particular, not so much for its meaning but for to whom it is applied is, I think, important. We are used to, in this context, hearing infertility described as disease. That it is is, of course, questionable. I, for one, am very healthfully happily infertile at this age and I would be worried if I got pregnant by some means.

On the other hand, am I infertile because I do have my ovaries. With egg donation, I could, as a matter of fact, gestate and have a child.

On the other hand, a thirty-year-old who is infertile may not consider this a disease at all because it is quite possible that she would never want to have children and, in her case, the infertility might be, as it is for me, a liberation.

So, just as an example of terms that I think deserve analysis in their own right, Alta alluded to the notion of family, the right to have a child, as distinct or separable from the right to reproduce and what does a right mean, anyway? The term preembryo--Professor Biggers has written a very useful and critical analysis of that term, quoting Margaret Sommerville's complaint that some of these terms, like preembryo, are used to be behavioral governing

rather than to tune in on an accurate scientific understanding of the terms and basically argues that here, and elsewhere, it is crucial to at least begin with a scientifically accurate term and to recognize that changing those usages may, as a matter of fact, be behavioral governing.

So I think all of these terms deserve analysis, and others as well. Another one that, to me, in any event, is important is one that is so commonly used, the term surrogacy which, obviously, suggests that a woman who gestates and bears a child, gives birth, sometimes, as one who is also the genetic parent, is not, in fact, a mother but only standing in the place of one who is.

Those are just examples of the ongoing need to be really careful about our language. It reminds me of a favorite quote that I have from Ludwig Wittgenstein who says that, "The essential task of philosophy is to overcome the bewitchment of our intelligence by means of language." Overcoming that bewitchment of language in this field, I think, is crucial from a policy standpoint.

[Slide.]

Let's talk a little bit about that underlying principle that I mentioned. As I said, there are different conceptions of justice. Justice is another term that may treated ambiguously but, in common parlance, it is often equated with fairness, with equality, with equity. It is also, I think, very commonly considered different from sameness and equality is considered, I think, often, in common parlance, as meaning sameness.

But it doesn't. Just think of what equality means even mathematically. It means the same value. Arithmetical equations, or even algebraic equations, don't mean that what is on one side is exactly the same as on the other but basically means that we have the same value, despite different arrangements.

Obviously, I like the algebraic form because I can put my XX equals XY in there and we are saying that women and men are not the same but they certainly have the same value. I can expand that to express Turner's syndrome or Klinefelter's syndrome, which are different chromosomal arrangements but which describe people who, while chromosomally quite different, certainly have the same value.

So the conception of justice that I want to argue as fundamental in the development of regulation or policies about assisted reproduction has to do with this notion of attention to differences, differences as opening us up to possibilities of discrimination or injustice and, therefore, as demanding attention to see that that doesn't occur or that it occurs as little as possible.

[Slide.]

I am going to talk, in particular, just a little bit, about some major differences with regard to assisted reproduction, those based on class which we have said, I think, too little of today and those based on gender or sex, which I think we could say more of, both today and at other times.

Some differences, of course, are changeable, some not. Some are equitable and some are not. Justice requires that we at least try to reduce the inequitable impact of differences where that is the case, where there is no inequitable impact, "Vive les differences." Let's keep them going. They enrich us as a society.

[Slide.]

Obviously, there are gender differences that are relevant to assisted reproduction that have to do with the fact that it is mainly women's bodies that are affected by whatever interventions are undertaken but also affected by the gender factor, that women still do tend to be the principal caregivers of children who are born as a result of assisted reproduction and, in general, the principal caregivers of those who need care.

[Slide.]

This is an aspect of a gender difference that a medical student whom I had some years ago, a colleague of someone who is sitting in the audience, checked out and did a little study on, a recognition of the fact that, because of assisted reproduction, women can be genetically or gestationally related to offspring with a separation between the two, gives rise to the recognition that biological ties, for women, are at least of those two separable sorts whereas biological ties for men to their progeny is only of one sort.

So the question of whether, for women, themselves, whether in the context of assisted reproduction or not, the gestational time may be more important than the

genetic is one that asked to a group of women coming to our clinic--not to the infertility clinic but women of reproductive age who just came to a clinic and were asked--I won't go through all of the questions that had to be asked to get up to this part of it--that, if they could only--if they wanted to have a child and the usual route to pregnancy didn't work, the usual recipe didn't work, and they could either be pregnant and give birth without being genetically related or be genetically related without being pregnant and giving birth, which would they choose.

[Slide.]

We also, in that same study, asked the question of men about their partners. Obviously, they could only be genetically related to a child, but which would they prefer if they had their d'ruthers for their female partner. Which would you choose, assuming your partner is open to either alternative? Your partner would carry the pregnancy and give birth without being genetically related or genetically related but not be pregnant and give birth.

[Slide.]

What was interesting in this study, and even if the numbers had come out very differently, it seems to me

the very fact that that question can be asked is important in terms of counseling and in terms of appreciating the way in which women and men regard having a child biologically was that we had an insignificant majority of women, a statistically insignificant majority of women, who said gestation was more important to them than genetics, but a significant majority of men for whom having their partner's genetically related to the offspring was more important.

That has some implications, for example, around the possibility of a carrier couple, a couple who are carriers for an autosomal recessive disease, having the possibility of eliminating the one-in-four risk of every pregnancy of having an affected child, and yet having a child who is biologically related to both by having egg donation with the sperm of the carrier male and the carrier female carrying to pregnancy, eliminating that one-in-four risk.

But, in general, it also suggests what I think is an important gender difference for counselors to take account of, namely, the fact that infertile women experience, in many cases, a double whammy. The loss of the opportunity to ever bear and give birth to a child is

an additional loss to that of never having a child who is genetically related.

[Slide.]

Some class differences relevant to assisted reproduction. I think we are all familiar, but we haven't paid a whole lot of attention to it today, maybe because we are so familiar with it, that infertility treatment is not an option for poor women, in general. The under allocation of the technology for those who cannot pay or have it covered is very, very clear.

Even when one carries that out to other context, even situations in which some infertility treatment is given, the kind that is given to women who are not as economically able as others may be compromised for that reason. When we think of the cases of very high-order multiples that have been born, the McCoy septuplets, for example, the Chukwu octuplets, those were both cases in which we didn't have any fancy reproductive technology that was provided. We had plain old Pergonal or Clomid, doses that produced those high-order multiples that could have been monitored in those cases more carefully even in those

instances by the generalist obstetricians who provided the drugs.

But, in the possibility that those couples had been more affluent, most likely, that greater monitoring or the possible use of assisted reproductive technology would have taken place.

But another possibility to consider around class differences in assisted reproduction is the possibility of over-allocation of the resources, that those whose income or coverage level brings or increases pressures to seek treatment because they can pay for it and because it is there, that they are, therefore, impelled to try to obtain that possibility of having a biologically related child.

Then, of course, in general, the notion of third-party involvement which usually does involve a discrepancy--Alta, I think, alluded to this--between the income level of donors and that of recipients. With my students, I remember having in front of me, during class, the announcement of supposedly the world's first egg-donor program at the Cleveland Clinic. One of my students signed up immediately. It was going to $900 to $1200.

About three months into the program, I said to her, "How is it going?" She said to me, "Hmmmm; if I weren't a poor student, I would never be going through this." Granted, she wasn't really poor. But, had it been ten years down the line when she was a whole lot better off financially, she would hardly have volunteered for that particular program.

[Slide.]

I think a very commonly supported framework for assisted reproduction could be called libertarian. It is a framework that generally stresses individual autonomy or choice as paramount implying that the primacy of procreative liberty or reproductive rights should hold sway over anything, the parents' right to have a child. John Robertson has strongly argued from that perspective, for example.

A limitation of that libertarian understanding of justice in a capitalistic society such as ours is precisely the one that I alluded to in the last few slides, namely, that it really applies only to those who can pay or are covered. In other words, they are the ones who have

reproductive freedom, the ones who can pay. Those who can't, don't.

It also implies, I think, problematically, that the right to a biological child is somewhat comparable, at least, to the right to property, that I have a right to have whatever I can pay for.

[Slide.]

I think these are some steps, if we are going to pay attention to differences that inevitably arise in these cases, in these questions; we have to avoid a tendency that has become very predominant in our society of being politically correct in our use of language by using couple language, for example, as if partners are equally affected by these decisions, by using other terms that seem to disguise or mask real differences that are class-based or gender-based or any other based, that we need, instead, under the aegis of justice, to pay attention to differences, to identify them, whether they be based on gender or class or race or sexuality or anything else.

We need to then determine whether they are associated with inequality because, if they are not, then that is fine; we don't have to go any further. If they

are, and if they are changeable, then it seems to me that we have to find ways to change those differences that are associated with inequality and, when the differences, themselves, that are associated with inequality are not changeable, then we have to at least make efforts to introduce measures that are going to reduce the inequitable impact.

Socially, we certainly have an example of that kind of effort to reduce the inequitable impact of differences that can't be changed through the Americans with Disabilities Act. That is a government mandate, recognizing that some things we can't change. We can't make the blind see or the deaf hear, but we can certainly do things societally that will reduce the inequitable impact.

[Slide.]

These are some of the suggestions that I think ought to be in place if we were to develop a truly egalitarian understanding of justice in assisted reproduction. We would have the same standards for research and therapy applicable to all regardless of income.

We would have regulations that are applicable to private as well as public sector. If something is demanded by justice in the public sector, it also ought to be demanded as justice in the private sector, and vice versa. We would try to develop regulations through the participation of those most affected. That is in order to overcome the inevitable nearsightedness of those who haven't experienced what the loss of fertility is, or what treatment for infertility that produces failures in that effort entail as well.

So it seems to me to overcome that inevitable flaw among groups of policy makers is that we have to really attempt actively to involve those who are consumers of the technology in the development of policies.

Some years ago, I served about five years on the Army's Breast Cancer Research Integration Panel. The involvement of consumers in that assessment of proposals and the determination of awards was a radical step for us to take in that context, but one which gradually not only the panel participants but the investigators, themselves, who submitted proposals generally supported.

That brought input that was relevant and that would not otherwise be there.

[Slide.]

I want to really just touch briefly on two areas of research--Andrea touched on the ova--well, she really touched on ova donation rather than ova freezing--as other areas that I think are worth discussing from the standpoint that I have been developing here.

Basically, I would argue for support of the research in these two areas on grounds of their potential for reducing some of the gender inequities associated with reproductive technologies. That doesn't mean that there aren't other possible problems that are raised that I think have to be considered, but, at least with regard to this one aspect, embryo-splitting obviously allows some women to have a chance at fertility that would not otherwise have it, while many women, of course, have superfluous embryos, superfluous ova and then superfluous embryos.

Some women don't and embryo splitting allows them--ova freezing, which, from a gender standpoint, allows women to, on the same level as it would allow men although men are not as interested in it as women since their

capacity for providing sperm extends so much farther into their life span, but it is a match with their capacity for providing sperm at any point which allows women to preserve ova for later use.

It also, I think, suggests the advantage, if it is an advantage, that if women choose then to become pregnant later when they would have greater genetic risks, they would not be as likely to involve or need, certainly, ova donation, third-party involvement at that point. So, narrowing that family framework and avoiding third-party involvement in reproduction seems to me to be an advantage for them.

So there are, in general, advantages that reduce some of the gender discrepancy for women that I think support moving ahead in those two particular research areas. As I said, and we can certainly talk about them during the discussion period, there are still some concerns about the potential that those technologies involve, for example, based on age and genetics, for contributing to less traditional or distorted notions of family relationships, identical twins born years apart, or even generations apart, for example.

[Slide.]

To conclude, though, I want to just put together some of the elements of what I have talked about that I think could be components in the policies or regulations that would ensue from those considering them on assisted reproduction.

The need for clarity and accuracy and consistency in the use of terms, right at the outset, any policy document and any discussion. Someone alluded to that today, the use of words by clinicians as distinct from researchers as distinct from patients but even as distinct from one patient to another, that that has to be laid out, that the question, what do you really understand by that, what do you mean by that, gets recurrently asked is very important.

The encouragement of adoption as a route to parenthood. It seems to me there is a compelling argument, and that is why I put it right in that recipe book at the outset for medical studies who may have a problem years from now that they want to have a child. If we are going to credit the notion of parenthood as more than biological, as much more demanding than simply that gamete or even

gestational component, then the encouragement of adoption as a route to parenthood can be effectively rendered by physicians as well as society at large, greater support for infertility treatment for those who can't pay with caveats that I won't go into right now because it would not be arguing that there ought not to be limitations in general, support for research on embryo splitting and ova freezing.

[Slide.]

Here is one that I didn't get to that I wanted to spend a little time on, the option to dispose of extra embryos without direct killing. I must have missed a slide earlier on that I won't try to go back to right now. It has to do with another term that is commonly used that deserves analysis but has been very controversial for a long time in the infertility-treatment area, and that is the term "disposition" of extra embryos, that problem.

What does disposition mean? It means placing something. There are various placements that are possible for extra embryos. We know that one is to donate or to transfer them to another women. We know that another is freezing, a very commonly pursued option. Another one is

use in research. Another one is killing. And another one is letting die.

Now, those last two are commonly talked about as destruction of embryos. If one of the differences that we want to keep in mind in this pluralistic society is that we respect differences in moral positions that different people have, it seems to me that distinction between killing and letting die, which is commonplace in ethics and law in general medical practice, is one that we could bring back to apply to those embryos for whom some people have no problem throwing into a refuse container but other people do.

Our suggestion is that practitioners, themselves, recognize that possibility and at least offer to people who have that reservation about living embryos being thrown away, that there is an analogy here with letting people die when they are probably dying and when, to keep them alive, we would have to make them undergo extraordinary and optional treatment.

Every day in our hospitals, we do allow people to die when they are in that condition. A single embryo or extra embryos in a Petri dish are, in a sense, more

probably dying than that they have the expectation of becoming clearly a person.

So I think it is at least possible that one can, if one buys into this, recognize that for individuals for whom respect for extra embryos makes them troubled about the idea of throwing them away, or discarding them, we can allow these embryos to die on a Petri dish. We can forgo that extraordinary treatment that would otherwise have to be given and, if you wish, you can even bury them, as some people bury limbs that have had to be removed with the body. Just respecting that difference in moral position, it seems to me is something that can be done even now and would be a way of addressing that general plurality of different moral positions that we have in our society. That is what I am getting at and I had a slide for earlier on that you won't see.

But the application, in general, my next suggestion to private as well as public sectors of the same regulatory standards, what is right for one group seems to me ought to be right for the other group. The recognition of innovative treatment as a subset of research--Andrea talked about that more at length and I wish I had more time

for it, but I think that is where it belongs. It is a subset of research.

The involvement of those most affected in the development of policies about assisted reproduction so as to overcome the inevitable near-sightedness of those who are not involved as affected either because of successful treatment or failed treatment.

[Slide.]

I am going to conclude with another cartoon. "Where do we come from? In my case, it was in vitro fertilization from the sperm of a deceased male. How about you?" The other guy says, "I think the stork theory has seen better days." That was a very old-fashioned recipe that never worked and wasn't exactly honest.

Let's hope as we discuss alternative recipes, we are a lot more honest and a lot more effective. Thank you.

DR. BORROR: So we are going to pause to take a break now and we will meet back here at 4 o'clock.

[Break.]

DR. WOOD: We will go ahead and get started. We are now going to begin the second half of our panel which includes three speakers and then some time for open

discussion. I need to make a housekeeping point; we really do need to be done and out of here, on the way out the door, packed up and going at 5:45 because apparently anyone who is not supposed to be on the NIH campus turned into pumpkins at 6:00. Alarms start. Gates close and you are here forever.

I know our speakers will keep to their times and we will have discussion but we do need to keep this moving forward.

Again, I am from the FDA's Office of Women's Health. FDA, obviously, has a big interest in all of these discussions but, the Office of Women's Health also has an interest in understanding both the scientific and clinical ramifications for the women as well as for their offspring and then, as well, what are the women's perspectives when it comes to a number of the ethical and legal and policy implications as well as the scientific and clinical questions.

Several of our speakers this afternoon will address some of these questions. Our first speaker this afternoon is Cindy Pearson who is Executive Director of the

National Women's Health Network to provide the women's health perspective.

Women's Health Perspective

MS. PEARSON: Good afternoon. Thanks. Not only am I lucky enough that I don't have to use powerpoint so you don't have to see me struggle through this recalcitrant computer like a lot of other speakers, I am also going to make just very brief remarks, so I am going to get us way ahead of this "get out by 6:00 or turn-into-a-pumpkin" deadline that we are facing, partly because I have to leave even a little earlier than that, which I am sorry about. I want to miss as little of the discussion as possible.

As Susan said, I am with the National Women's Health Network. That is a group that is very well known to a lot of FDA folks but maybe not so well known to the basic science and ART practitioners in the room. We are a twenty-seven-year old health advocacy consumer watchdog group.

Susan has introduced us as talking about the women's health perspective which is what our members send in their dues every year to have us do. But I want to acknowledge that I don't see the women's health perspective

as completely distinct from the patient perspective or even, to some extent, to the provider perspective. We do have issues that probably have unique nuances on this subject but we don't see ourselves at all in counterpoint to women and men who are considering or going through or have gone through infertility treatment.

Now, I want to just reflect on what brought us here. The title is Social, Ethical and Policy Issues of Evidence-Based Assisted Reproductive Technology. It seems pretty clear that the subtext that has sort of emerged out into the actually spoken text is that we have got regulatory and issues pending and we have got the need for more research or the desire for more research that we are probably the catalyst to get the people who did such a good job of organizing the meeting to spend the time and get access to the resources to make this happen.

So, I am clear that that is probably where my comments and the comments of the National Women's Health Network can be most useful because that is what everybody is thinking about, should regulation change, should things that are on hold be finalized and what would be the most useful way for that to happen.

Similarly, with research, we have tried to do our fair share of getting more money for the NICHD to use for good research projects on women's health and we would like to see more in this area and so what specifically would we be interested in seeing.

We, also, as a consumer advocacy women's health group, have a perspective that includes probably issues that are broader than what anyone expected to consider here in terms of social issues. I will touch on them briefly, but with the recognition that, while they may interest people here as individuals, this probably isn't the place that can make the changes necessary.

One that has been commented on throughout the day of the restrictions on federal funding for embryo research would obviously--we consider that a women's health issue. While we agree with the speakers who have said there is zero chance of that changing in the near future, it is something that we could continue to describe as part of the women's health needs in this whole arena.

Let me just address what we would say is a women's health perspective on the questions we are talking about today. Our context is that the women's health issues

related to infertility and assisted reproductive technologies balance women's right to know and women's right to choose.

Women's right to know, in terms of assisted reproductive technologies, are the obvious rights that many people have talked about today; the right to know how effective a procedure is, the right to know what it will entail and what it will cost, the right to know what is known about short-term risks of the procedure and the right to know what hints there are at possible long-term effects both for the woman, herself, and for her offspring and, in the offspring, the right to know as much as is known about some of the seemingly very good outcomes for the offspring of the first wave of assisted reproductive technology.

Also, we believe that the right to choose is an important women's health issue in assisted reproductive technology. The right to choose, of course, is the right to choose if a women, herself, or as part of a couple, wants to have children, when she wants to have children.

When we are talking in the childbirth arena, we talk about the right to choose the setting of her birth and in the infertility and assisted-reproductive setting, we

talk about the right to choose what level of assisted reproductive technology she or the couple is comfortable with. That leads right to one of the things that we bring up, we feel is a need to bring up as women's health advocates, is neither of these rights has been made fully available to women in the United States.

You could tell I could almost not quite finish my sentence about the right to choose assisted reproductive technology and choose how far along the pathway the sort of suite of options within assisted reproductive technology a woman is wanting to go without reflecting immediately on the enormous barrier there is for women in the United States in effecting that choice; that is, as other people have mentioned, it is a choice constrained by class and financial resources.

I want to make just a short mention of an area which is beyond the scope of this meeting but we see as a women's health issue is the more recent, just in the last five years, example of welfare reform and policies creating barriers to women's right to choose whether or not have to children and how many children to have.

Now, no one would describe welfare in the United States as a support for assisted reproduction but it certainly was thought of a support for assisted family survival for families who didn't have the financial resources for women and their children, who didn't have the financial resources to keep their children fed and housed.

That is now not guaranteed as policy of the United States government after generations long, although crummy, but a right of women to turn to the government for assisted family survival. That is now not guaranteed either beyond a certain time period or beyond a certain number of kids in some states.

So, in the same way that many of us today have reflected on the issue that class and economic resources pay for the ability to get assisted reproductive technology, and I would say that, as a women's health advocate, that is an issue that we need to work on to rectify. Similarly, we think it is important to look in the bigger context and see the full scope of policy impact on women's abilities not only to have children but to, then, maintain their families.

People have also mentioned another barrier that there has been for women's right to choose assisted reproductive technology and sexual orientation. That is a concern we have also, as a women's health advocacy group, that rights should be inherent in the person and not constrained by other people's beliefs about sexual orientations and impact on someone's ability to parent.

Turning to the right to know, I described briefly but I will just restate, what do we think is entailed in women's right to know. It is the right to know what is experimental and what is more well established. We acknowledge, as has been emphasized today, that, in this field as much as any other field that we deal with, and being a generalist women's health group, we are active on cancer issues and contraception and menopause, this field has the fastest changing boundary between established treatment and experimental treatment of almost anything we deal with.

That poses a real challenge, I think, in the effort of very well meaning clinicians and researchers to give women seeking services a fair description of is this something that we can describe as routine or are we on the

frontier, as someone used that term earlier today that I liked.

But we do believe, from the women's health perspective, that this is a right women have. The practitioners just need to strive to do the best they can in any month, in any cycle, to describe what is now more standard and what is still experimental.

What are the known risks? I am sure everyone in this room is very committed to sharing what they know about the short-term impacts and what risks there may be and what is the preliminary information that we know, which may not be actual cause-and-effect information, may only be association. But what is the preliminary information we know about long-term effects.

We have heard already that there is tension in the clinical setting and I guess between the clinicians and the researchers of how much to put in a consent form, how much to put about information that may not be cause and effect and may be kind of scary.

I will just give you a voice from the women's health community; if you know, we want to know, too. It is true that a consent form can be written in a way to really

intimidate and frighten people, but a consent form can also be written in ways that are empowering and there is a whole world of people who research the process of doing research, and some of them specialize in the informed decision-making process.

So that resource does exist and maybe there needs to be a little cross-talk between those two groups of people.

Then, what is effectiveness? I am sure all of you are familiar with this history of the pressure from the outside that probably met with some like-minded support from the inside of the service-providing community to give information to women about effectiveness in a way that was useful in the terms they wanted which, for most women, is what chance do I have, walking in here, ending up with a baby in some reasonable amount of time in the future.

There have been tremendous strides in the last decade of making that kind of information more available to women, but it was interesting hearing, even today, still some back-and-forth between people of, "Well, it depends on how you describe the statistics." So that isn't a solved

problem is, I guess, what I would say. There is much better information available.

I have the impression that women are much more likely to get the information in a way that is useful to them but don't rest on your laurels. Keep it up and keep working at that, I guess, would be one piece of feedback I have to this community.

Then I want to bring up an issue that I know is of concern to the provider community and to the women and couples who are considering or going to through treatment now, but it is also a concern, and possibly balances out a little more in our near ground rather than distant ground, of the women as donors in the women's health community. What are the women's health concerns for women as donors?

This is where the dilemma that is probably pretty well sorted out for women who are patients becomes more of a dilemma, and that is that women do have the right to choose based on incomplete information. It is all they can do. While we, as a generalist women's health group, will argue that healthy people shouldn't be encouraged to choose potentially risky interventions to reduce their risk of developing disease in the future until things are really

well sorted out--and I point to HRT as a recent example--we accept and embrace the idea and the reality that people who have conditions now, particularly serious conditions, have to make decisions about whether they will accept a certain treatment based on somewhat incomplete information.

We believe that infertility treatment is like to that, that the steady drumbeat of time and those monthly cycles going by put an urgency on the woman's decision about accepting treatment that means that she is--many women in that situation will likely be willing to make a choice based on incomplete information.

But for women as donors, it just seem more difficult. They don't have a drumbeat of time except to the extent that they are motivated by altruism. They do have an incentive of reimbursement. The tension around making a reimbursement that adequately recompenses women for their time, which is significant, and for their discomfort, which is significant, but holding back from going so far that the payment is, in and of itself, an incentive seems to be one that is not solved. It is not a solved problem.

All of us who work with interns or who work with college students here at least jokes, if not more serious reflections on, "Whoo; that money would really help and should I maybe do it because that money would make a difference," and maybe I have some altruistic feelings that this would be a nice thing to do, too.

So those are the issues, I guess I would say, that we bring as women's health issues. To sort of bring it back down to a focus that is useful, in terms of the policy issues that we are asked to reflect on, the research issues that we are asked to reflect on and the regulation issues that may be pending, obviously, from the women's health perspective, the role of research, our role, from the women's health perspective, is to get more money for you, get more resources available to the community as a whole.

But the long-term effects, the effects on donors, the effects on children, offspring, I don't think we have another venue to which to turn outside of this setting, the people who have worked so hard to make research resources available up until now and those of you who have devoted a lot of time. Aside from your compatriots in other

countries, we are looking at you and this is what we need from you and we need more of it, and we hope we can support you to do more of it.

Similarly, the role of policy, I think, has an indirect effect on getting women continued good information about effectiveness and a continued commitment of the majority of the practitioners to be modeling good behavior in terms of very high quality informed decision-making processes and informed consent.

Those are difficult to regulate specifically, and regulation isn't what is making them happen now, to the good extent that they do happen now. But I think that policy efforts on the large scale have led us to where we are and that is important.

Specifically, the regulation, obviously we would agree that that safety, sort of bottom-line safety, of regulating a requirement that decreases the risk of communicable diseases being transmitted as part of assisted reproductive technologies is important. It seems insignificant compared to the many other questions, but it is sort of a bedrock.

I don't think that the women's health community is convinced that regulation of the genetic aspects of assisted reproductive technology is really sorted enough, or mature enough, or clearly evolved enough, to where we can see a specific women's health need that would be met by that kind of regulation.

I am sure that its not quite as clean and crisp a comment as people would like, but I think it is a reflection of the state. It is evolving societywide and within the women's health community. We continue to discuss it amongst ourselves and evolve our own positions.

So those are brief remarks. It is late in the afternoon. I look forward to the discussion that we can all have together at the end of the afternoon, if we can keep our eyes propped open long enough. Thank you.

DR. WOOD: Thanks, Cindy.

Our next speaker is Pamela Madsen. She is the head of the American Infertility Association. She is going to be providing a patient advocate perspective.

Patient Advocate Perspective

MS. MADSEN: Whenever I give these kinds of talks, when we are focused on infertility and the infertility patient, I always feel like Exhibit A.

[Slide.]

Okay; here I am, a fertile woman, patient advocate for fifteen years, fertile through ART, mother of two IVF children, one Birdseye, which I call for frozen. He doesn't wear mittens when he plays in the snow. He is very familiar with the cold. I always feel, among the science, a voice is here to say, Yeah; guess what, folks? It is not a word.

[Slide.]

Infertility is a disease.

I suspect that all of you knew that, but we have played with the language a little bit. So let me bring it back again as a patient and as a person who advocates for people who are suffering from this disease that affects one in six. So somebody's sister, brother, neighbor, friend is suffering right now with infertility.

It affects men and it affects women and it affects couples together and whether the infertility began in the man or began in the women, ultimately, it could be a

combination and belongs to the couple. Yes; there are single people who are infertile and who are trying to build families. The American Infertility Association supports them.

So how does it impact on our lives? I promised I wouldn't go too much into the psychological or the emotional aspect of infertility but I would be remiss if I didn't say to you, don't tell me that we don't warrant treatment, we don't warrant risk taking, because infertility won't kill you. Infertility won't end your life.

My response always to take a breath. I look out and I say, no; infertility does not end your life but it can stop your life. It can prevent a person, a couple, who is experiencing infertility from attending family events. It can prevent us from going about many, many of the ordinary situations that all of you go through every day, we often stop.

So, no; it doesn't kill us, but it can stop us. When we talk about should we take the risks for treatment, please look at us, what happens to us when we don't take the risks for treatment to treat our disease and have a

child. And, yes; infertility affects poor people and it affects middle-class people and rich people and white people and black people and Hispanic people and Indian people. All kinds of people all across the world are affected by infertility.

Yes; access to care is an issue. Oftentimes, you are only going to see people with resources in clinics. Not always rich people in clinics, people with resources, people with credit cards, mothers, lucky enough to have a house to mortgage, people paying out of pocket even though they are working people and have health insurance and find out that somebody in the health-insurance company also somehow thinks that may infertility isn't a disease and isn't terminal so, therefore, shouldn't be covered.

So just because you are just seeing a certain kind of face in the clinic doesn't mean that there aren't lots and lots of people out there who want to have families and who can't because of access-to-care issues.

[Slide.]

So here we are. Why are we here? Why now? I think it is because infertility, ART, is coming of age, just like Elizabeth Carr who is the first IVF kid in the

U.S. who is now a college student here in Boston. She has come of age. So has ART. Now, we are beginning to question what are we doing. We are looking back. We are seeing where we are going.

Why are we doing it now? Because enough time has just begun to pass for us to stop and think and contemplate. I think that is a great thing. I'm glad that we are all here and that we are all talking about this. And many of us, including myself, are just reaching the years when maybe other kinds of health problems could be showing their faces.

I started treatment in my twenties. I am in my forties now. And, you know what? I am curious about what could be in my future. When I look around with other people who have completed their treatment and have called me up and asked me for answers, and asked me about research, about women who have completed their treatment and where their health is now and is there anything they should be worrying about, I kind of have nothing very much to say because we don't have good solid research about the long-term effects of infertility treatment.

What would I like to tell them? I would like to tell them that there is great research with lots of data and the news is good. And I would like to reassure them because I feel pretty good. My friends look pretty good. But we don't really know.

[Slide.]

I believe that the time for the research may have been yesterday but it is certainly today and it is certainly now.

I was asked to come up with where I see, as a patient advocate, the gaps in research are becoming apparent. I mentioned earlier, I am a mother. I a mother of two IVF kids. You know what; they are not babies anymore. They are ten and they are thirteen.

When putting together an issue which I gave to you folks, our children for our membership, and we went looking for information and data and research to give our membership about our children, I was shocked how little there was. Folks, that is a bad thing. I am not saying it as an alarmist because I think these kids are perfect. These are cute, smart, funny kids who look pretty good to me. But I don't have the research.

I can't reassure people or warn people except my own life experience and small studies. But I am not sure how good they are because they very small and they are selected programs and selected information and there is no large database of our kids and their information and their health.

As a patient, as a patient who speaks to other patients and as a mom, I want the meat. I want to know how these kids are doing. Is it the fault of our doctors and researchers that we don't have it? No. We are very young field. As I said before, our kids are first becoming of age.

In the beginning, there was just IVF babies. Now we have kids an we want to know about them. There are a lot of kids. There are a million kids. In France, they put together a national database of IVF children. It is not done here yet. I loved the lady in the back before who I think was from--I don't know if she was from NIH--and who said, "We need to work together as a community and put the money into this and have the patient advocacy organizations and the professional organizations work together to put together a registry of these kids," so, for the next

generation of IVF parents and prospective patients, we have real information to tell them instead of me just having to tell them how smart Tyler and Spencer are because I don't think that flies.

And you know what? Spencer is a cryo kid. I mentioned that before. So when I was putting together my great issue on our children, I went looking for a study about cryo kids. And they froze my embryo. How many of you have had your embryos frozen? And they put it in. Nine months later, I gave birth to this beautiful little boy. And that is really all I know, that the beautiful little boy started life as a frozen embryo.

As far as I know, there is no single study out there about cryopreservation of the human embryo in long-terms studies of these kids. I would like to see one.

ICSI kids; we are talking a lot about these ICSI kids. Yes; I think we've got it now. We need to see more about the ICSI kids, more long-term studies, bigger data pools. I would like to see some psychological follow up on donor kids and donor families, donor-egg kids, donor-sperm kids and now that it is going to be federally funded, donor-embryo kids. We may have a lot more of those. So I

think that we need to look at some long-term studies, some psychological studies, how these kids are doing.

I think I already answered, why these gaps? What is there? Surprisingly little. I think I answered the why. I think it is really because it is so new. We are such a young field. We are just beginning to get to the point where we are looking for the information and, for whatever reason, whether it be funding that has been scarce for this type of research, it has not been done.

I think that because funding for infertility has been in the private sector for so long and funded really through patient dollars, that our urge, as patients entering the system, was on conception. It was only until later that we are deciding that it is more than about conception. It is about, then, our kids, our families and the long-term health.

[Slide.]

So what about me, the infertile woman, the infertile man? What about me? Where should research dollars be channeled for me? What do I want to know? I want to know if I can separate my exposure to infertility

medications, gonadotropin therapies, from my infertility. What does that mean?

I may have health problems because I wasn't a fertile woman. Sometimes, we don't really kind of talk about that, the fact that we are dealing with a population of people that have a distribution called infertility which may lead to other kinds of reproductive health problems down the road, and how do we separate the fact that we have been exposed to all this different kinds of stuff on our bodies from our infertility.

There have been a few studies and they have been flawed. They haven't been, in any way, reassuring. Some of them have been frightening, and they have been frightening and flawed. I would like to see some good studies done that would help patients understand if taking fertility medications will further compromise any health risks they may have as an already infertile person.

I want to know, do I really do better as an infertile person if I do conceive, because there has been some research, some little theories out there; well, because you are infertile and if you don't have children,

that is really the issue. That is really why you are going to get ovarian cancers because you are not having the kids.

If we get you pregnant, that is going to help you avoid breast cancer and ovarian cancer. I am sure some of you have heard some of these studies. I don't know how good these studies are. I would like to see some good studies around this.

And, if that is true, that if I do conceive with fertility medications on a healthier person, what happens to me if I just expose myself to more medications and more technologies and I don't get pregnant. What kind of shape am I in then?

And research on egg freezing. We need to learn how to effectively thaw eggs. We need to learn how to do that. That is really an important thing to women. We need to fund that. Why? Well, there are some women who are exposed to chemotherapy at a very young age who would like to be parents one day, who would like to be a mommy.

It is very basic, you know, wanting to be a mommy or a daddy. That technology will help these women who are dealing with cancer, who have enough to deal with with that loss and that fear and then also have to grapple with, at

the same time, maybe at fifteen, that motherhood may also be taken away.

And, yeah; it's true. We are naughty girls. We are postponing childbearing. We are getting college educations, just like the guys. We are getting careers. And we may be waiting longer to marry and have children. And maybe egg freezing may help there. I am not suggesting that all women should go out and bank their eggs and then thaw them and use that later in life. But it does create an option for some women who might find that really helpful. I think that we really should put some dollars there.

[Slide.]

We talked a little bit about the genetic basis of inherited infertility and what does that mean. I think that there should be some research dollars put there. And I wrote down endometriosis. But PCOS. There are a lot of underlying diseases for our infertility that involve chronic pain, that really affect quality of life. PCOS, polycystic ovarian disease syndrome; most of you are scientists and doctors. Heart disease, diabetes.

I almost said it is not just infertility. But I didn't mean it in a way that diminished infertility. But a lot of the diseases that cause infertility have other ramifications on our lives.

[Slide.]

And, yeah; I do want to get pregnant. I don't really care if you are going to help someone ten years from now and it is going to be a whole lot safer for them in ten years because, you know what, I am selfish. This is about my life. I want to have a child. I want help and I want help today.

I need the research done today. We have not solved the problem associated--again, I will go back to the aging woman's egg. Again, we are not talking about women who you would consider old. Surprisingly young women have ovarian aging issues. We talked about FSH. That is a marker that we have been using to help women understand whether or not they have ovarian aging.

These women call me daily. This is not a small problem, ovarian aging. This is big. They want to know what can I tell them, what is the newest treatment out today. For way too long, I have had to tell these women

that I have nothing more to tell them. There isn't anything new. There aren't human trials today, if they wanted to volunteer or participate in, that they could participate in.

They are doing great work with mice. They don't care about mice. They want to know. They are not scientists. They are not regulators. They are people suffering from infertility who want help now.

[Slide.]

Yes; we want fast-tracked research. I put it all by itself. But I should have added another button. We need money for that fast-tracked research. We need it now just from the private sector. The federal government is getting involved with us. That's great. They want to provide oversight? They want to have input? It would be nice to see funding. We need dollars.

[Slides.]

I was going to spend a little time on this but I am not sure that I am going to. As a patient advocate, I listen a lot to what the doctors have to say about all this. I think I will just click these through and you have them in your book.

[Slides.]

And I think you know what the regulators have to say. We have been spending a lot of time on this, so I am not going to waste your time.

[Slide.]

So I will tell you what I say. What do the patients say? What does the American Infertility Association say? We do understand that, to a large extent, we pay for the services that propel development in reproductive technology literally out of our pockets. Many of us are willing participants in experimental procedures but we do want the right to honest forthright information before giving consent.

I think that we are getting that. I think that our professional societies and our physicians and our researchers take informed consent very seriously. As a patient advocate, I can tell you the news is good on the home front. Patients are being consented.

We want to know what the doctors think might happen, what the possible pitfalls are, what the best guesses are, what our researchers and our doctors know and what they don't know. The urge to have a child is

incredibly strong and many people--well, Louise Brown's mother took a tremendous leap of faith in her desire to have a child.

Patients who suffer from a disease that is life-stopping often, whether it is infertility, cancer, any disease that has to do with life, beginning of life as infertility is, or ending of life, often want informed consent. They want to know, but they don't want the decision to take the risks taken away from them. They really don't.

[Slide.]

As a patient advocate who looks over this field and listens to what the doctors say and what the regulators say, we look at the IRB process and we don't know if there is a way to overhaul those, to provide more oversight, rather than reconstruct a new system, rather than put an IND application into the mix, if that is really going to help us a patients.

We worry that it has stopped research now. It hasn't slowed research. We talked about permissive. I heard the word--I am finishing up. It has stopped research today. But if the FDA is going to go ahead, what I say to

you is please, use restraint. Think about the money and the costs that are going to be passed down to the patients. Remember that this is about us today, not some other infertile woman tomorrow, our family today, our kids who we want to take to kindergarten on the first day, the kids that we want to read a storybook to.

It is about our families, our kids, and we have a very short window of time.

[Slide.]

So, on that, I will pop along because I have run ahead. But I love the fact that we are all together, that we have included the patient voice, we have included many kinds of wonderful thought leaders here today. I think that if we stay on that track, we are headed in the right direction in helping people who want to have a family have them.

Thank you.

DR. WOOD: Thank you.

Our final presenter today is Stephen Ory from the IVF Florida Reproductive Associates who is going to provide us with an ART practitioner's perspective.

The Practitioner's Perspective

DR. ORY: Thank you.

I would like to thank the organizers for putting this together. I have sort of the sense of being one of the blind men or blind people with the elephant. We have all been very busy working in very different areas and I think the opportunity to get everyone together to present what they have been doing with this problem has been very instructive. I have learned a great deal today and I hope that I can convey to you some of the activities that we have been involved in.

[Slide.]

I am here today representing the American Society for Reproductive Medicine, the Society for Assisted Reproductive Technologies and SART, and the Society for Reproductive Endocrinology and Infertility.

[Slide.]

SART and SREI are affiliates of the American Society for Reproductive Medicine with slightly different focuses. We are all interested in ART. ASRM is a large organization consisting of physicians in addition to obstetricians, gynecologists, reproductive endocrinologists, urologists, internists. There are a

large number of laboratory scientists, mental-health experts, nurses and other people who are interested in reproductive-medicine issues. We are also interested in aspects of the menopause and contraception.

SART has concerned itself exclusively with ART. There are 374 member practices in the United States and they represent over 95 percent of the ART programs. I will explain in a few minutes why those 5 percent that are not represented in SART are not part of that. SART has promoted advances and the development of practice standards in ART.

The SREI consists of 95 percent of the reproductive endocrinologists in the country. There are 870 of us. We have focused on research, education and clinical issues pertaining to reproductive endocrinology.

[Slide.]

The ASRM has a number of activities. I wanted to highlight a few that pertain to the establishment of standards of practice. Most notable, the guidelines for gamete and embryo donation which were revised this year is a lengthy document offering explicit recommendations or requirements, I should say, for screening for sexually

transmitted infections, genetic testing, psychologic screening, pertaining to sperm, oocytes and embryo donation.

The ethical guidelines have already been discussed this afternoon. They have direct relevance to the practice of ART as well and they have offered model documents for informed consent and have instructed us, in terms of proceeding with a number of new technologies, at least the ethical dimensions of those.

The ASRM Practice Committee has developed over twenty committee opinions plus technical and educational bulletins detailing a broad array of practice issues pertaining to ART. Our journal, Fertility and Sterility, has been published monthly since 1950 and it has addressed topical issues and controversies pertaining to ART.

The ASRM sponsors and annual meeting preceded by two days of post-graduate courses each year.

[Slide.]

There have been several allusions today to how we validate new treatment or how treatment progresses from experimental or investigative to being clinically validated. Really, it is much the same as it is in any

other aspect of medicine. We usually start with animal data. Dr. Bavister illustrated very nicely this morning the limitations of some animal data, and there are innovations and advances that we have not been able to test in an animal model.

Most of the time, advances have begun with limited clinical trials following IRB approval and an IND, when appropriate. These have generally led to randomized controlled trials at multiple sites. Much of the answers right now are coming from Europe because of, perhaps, an environment over there that may make some of this easier to accomplish.

But once this data is available, the ASRM Practice Committee with representation from SART and the SREI reviews the available data and endorses it when it is of sufficient weight to be found clinically valid.

[Slide.]

SART's activities have been a bit more focused in the SART arena. Since their inception in 1987, they have developed guidance documents which are essentially minimal standards for the practice of ART in the United States, and

they have been updated periodically and are current at this time.

In 1992, they received a contract from the Centers for Disease Control to collate clinic-specific data. SART has published the clinic-specific pregnancy rates since 1989 and has now been doing this with CDC sponsorship since 1992. They also oversee an inspection and accreditation of embryology labs with CAP and the ASRM laboratory group.

[Slide.]

For membership in SART, it is required that each member program submit the results of their clinic-specific success rate each year. These data are collected through September of the following year. They are collated and published about a year after the deadline. This allows us to follow all patients to the completion of they pregnancies so that those who conceive in December of the previous year will have complete pregnancy data in the following year.

All members have to agree to on-site validation and, if they don't, as is the case with Iraq, they are excluded or sanctioned in some other way. There is a

mandatory laboratory accreditation and inspection process. This can be accomplished either by the previously mentioned CAP ASRM process or it can be done through JCAHO or through the New York State Task Force.

All M.D. directors for the past several years are required to have reproductive endocrinology and infertility subspecialty certification. The lab directors must adhere to published standards and they must adhere to the published guidelines pertaining to ethics practice laboratory in advertising.

[Slide.]

There are several accomplishments that I think we can cite. By and large, the practice of ART here has been safe. There are effective drugs and techniques for providing it. Since the success rates have been published, we have seen an increase from 11 percent--these are the live birth rates--in 1987 to 29 percent in 1999. We will have the 2000 success rates in another six weeks.

Worldwide, over 1 million babies have been born including several hundred thousand in the United States. That is a very sizable constituency. And we have seen continued innovations including intracytoplasmic sperm

injury, preimplantation genetics diagnosis, cryopreservation of gametes and embryos and embryo hatching evolve over this period of time.

However, this has been accomplished in an environment with considerable social and political constraints. It has been done without NIH research sponsorship at least in the arena of clinical patient ART. We did not have the help of the best and the brightest. It has been a costly, inefficient, profit-driven process. I share Dr. Bigger's lament this morning. But it has been safe. In the U.S., there have not been any reports of sexually transmitted diseases arising from the practice of ART and it appears, in the U.S., at least, according to the registry data that we have thus far, which the limitations have also been discussed earlier, there does not appear to be an increase in anomalies that we are aware of to date.

[Slide.]

The unresolved issues have also been discussed today. I think the most important one that we are focusing on is reduction of the high-order multiple-gestation rate. It will be important to define and reduce the risk of preterm delivery associated with multiple gestations but

also, as was discussed this morning, which may be higher with singleton deliveries as well, possibly as a consequence of ART.

The risk, if there is increased risk with ICSI, has to be defined better. We need to define the role, the effectiveness and safety issues associated with PVD and we must preserve the future and ensure a mechanism of continuing to develop new technology and to improve on those that we already have.

[Slide.]

The challenge for us now was to balance the countervailing priorities of developing new technologies while preserving safety and efficacy. This requires flexibility in anticipation of new technologies in the climate of increased oversight and regulation.

[Slide.]

There are several unique features of ART in the United States. There is an absence of a federally subsidized healthcare system which encompasses infertility and ART. We do not have a national regulatory agency such as the Human Fertility and Embryo Authority in Great Britain or the Reproductive Technology Accreditation

Committee in Australia and, as Pam just said, the cost of ART is predominantly born by patients and they have very legitimate expectations of safety, efficacy and input into the process.

[Slide.]

There are a number of existing regulations of ART. I want to review just a few of these. This is a very selective depiction. CLIA '88 specifically addresses the andrology labs and all andrology labs that are involved with ART programs are affected as well. The FDA, as you know, has pharmaceutical and new-device oversight and they have recently asserted oversight of therapy with human cells involving transfer of genetic material. This impacts somatic-cell nuclear transferase, cytoplasmic transfer and coculture.

[Slide.]

The Fertility Clinic Success Rate and Certification Act of 1992, often referred as the Wyden Bill, was developed with ASRM, SART and RESOLVE sponsorship through Congressman Ron Wyden and specifically requires all of the ART programs in the United States to report their pregnancy success rates. The identity of each laboratory

affiliated with an ART program must reported and the certification of that laboratory has to be reported as well.

This is published and distributed and the individuals or the programs that elect not to respond are reported in the report as nonreporters. Since 1995, these results have been published on the Internet. In 1995, the 1992 results first became available.

[Slide.]

The Act also mandated the development of an inspection and certification process for embryology labs and that was the one that I mentioned earlier, which has been developed by ASRM and CAP. They required the development of a model program for the certification of embryo labs and their proposal for that was published in the Federal Register in 1992.

[Slide.]

The current status of the Wyden Bill is that over 95 percent of the approximately 370 ART clinics in the United States have reported their data. SART was awarded the contract to collect the data for the CDC. The 1995 results were posted on the Internet in '97. Thirty clinics

have had on-site validation in 2001 and, through the end of 2001, there were 85 clinics that had been certified.

There have been a number more that have completed that process. At this point, all of the clinics in the U.S. who are currently members of SART have either been validated or are in the process of being validated. There have been a number of programs that have not adhered to these requirements and have lost their SART membership which, at this time, is really the only sanction that SART can take.

[Slide.]

Other nonmedical agencies that have been involved with the regulation of ART include the Federal Trade Committee who monitors truth in advertising. They have fined programs that have misrepresented their success rates. The Occupational Safety and Hazard Act has jurisdiction over laboratory and offices regarding safety issues and has been active, state and local business licenses. The tort system, HMOs have all exerted various influences in the process.

[Slide.]

Genetic screening through the Department of Health and Human Services, the CDC, the FDA and Office for Human Research Protection all have oversight. For research in genetic testing and treatment, the NIH has jurisdiction.

[Slide.]

At this time, there are a number of things, and I think there seems to me to be an evolving consensus from what I have heard today, the screening and processing of third-party issues, standardizations of that would be welcome initiatives. We have standards in place that I have outlined that are effectively standards of care that better than 95 percent of programs in the United States adhere to.

Those that do not, we currently have no sanctions to take against them other than excluding them from SART membership. I am making a distinction between those that fail to comply with the standards of care from the out-and-out criminal activity, and there has been some of that. That is, then, completely addressed by some of the state statutes.

So it would be a welcome initiative to have some means of enforcing these practice standards and problems.

What we would not welcome are initiatives dictating the practice of medicine. I think Dr. Racowsky, this morning, really showed in a much more compelling and articulate manner than I can discuss in my remaining minutes, the difficulty in coming up with a strategy to reduce the high-order multiple pregnancy rate.

I applaud her and her group for the efforts that they have made but I think you can see from a process that requires considerable laboratory expertise, a large volume, and is now in its sixth iteration, that it would really be next to impossible to impossible to come up with national recommendations that will be applicable to all programs, just addressing that one aspect of ART, the number of embryos to be transferred.

[Slide.]

In conclusion, I hope that I have convinced you that extensive oversight of ART exists. We are not aware of any current regulatory model which is ideal. The ideal system, we believe, is one that would enhance safety while preserving flexibility in individual patient management and we feel that it is imperative to have input from all of the

participants in the process as your program today has accomplished.

Thank you very much.

MS. WOOD: Thank you.

If all the panelists could come forward, the ones who were here earlier included, Alta, Andrea, Mary, Cindy, Pamela join the table at the front.

While they gather, one other housekeeping note and that is to remind everyone that tomorrow's meeting starts at, yes, 8 o'clock in the morning. Please be here on time and ready to go.

Open Discussion

DR. WOOD: We have a few minutes for discussion. I guess I can ask the panelists if they have any initial response, but I did see Phil Noguchi had a comment.

DR. NOGUCHI: I can wait.

DR. WOOD: Did anybody want to make any responses to some of the presentations that were made in terms of reacting to each other, or do you want to jump in with the audience?

DR. MAHOWALD: I just wanted to make a comment. On the discussion of multiples, because that really is an

important one, but a comment that I might have made during the talk because, in discussing class differences, because the great majority of multiples don't come from reproductive technology, itself, but from infertility drugs which are more difficult to monitor but can be monitored.

Those fertility drugs tend, by and large, to be given more often by general practitioners rather than reproductive endocrinologists. So there, again, there is a class issue that is associated with that problem that needs to be addressed.

DR. ORY: I would agree with that comment to an extent. I am aware of the two cases that you alluded to in Texas and Iowa. Unfortunately, both of them were managed by reproductive endocrinologists--the one in Iowa was. I know the physician and I believe the one in Texas was, as well. Maybe not, but the point I was going to make is with appropriate monitoring, that risk can be reduced but we still do not have a management scheme that will allow us to eliminate the risk of multiple pregnancies with superovulation. In that regard, we are more successful with IVF.

DR. MAHOWALD: Monitoring follicle cells, even in those instances, could and should have been done and would have avoided the problem with the additional advice of avoiding intercourse during the cycle.

DR. NOGUCHI: First, I would like to just thank everybody for both this morning and this afternoon for your participation and willingness to not only just say how nice it is but to say what you like and what you don't like. Part of our job at FDA is to really do that balancing.

I guess what I have heard so far gives us some hope that actually we are talking pretty much the same thing with maybe the way FDA talks being somewhat of a problem and the concerns. What I can say is, and especially we will be establishing a new office this Sunday that will be overseeing all of tissues, gene therapy, cell therapies, the whole gamut of things. We are quite aware, and we are quite concerned, that the amount of regulation is balanced to the task at hand.

For ART, as we have been discussing especially today, what I would like to point out is that by and large, we think that the infectious-disease rules will cover 99 percent of everything. It is the 1 percent we are

really talking about. There, rather than talk about restricting innovation, what we are actually talking about is a third part of what FDA does which--Alta talked about safety and efficacy, but in our regulations, underlying the whole thing, is, first of all, safety to human subjects that participate in clinical trials.

The question that we are posing is, as part of that human-subject protection, we are not saying that human subjects should not take risks. But, as I have heard almost everyone say today, we want to know what information is available. We will posit directly that, for ooplasm transfer, there are many unknowns about it. We do know that mitochondria may be transferred. We have really no direct evidence that shows that the ooplasm transfer caused the pregnancy.

That is kind of the bottom line. If you are going to undertake a highly experimental therapy, what is known, what is not known, what will be gotten from the experiment being done at hand. It is really an informational exposure. It is a disclosure aspect that we are focusing on.

We do think absolutely that--or, let me put it this way. FDA learned, not too long ago, to be not so paternalistic. Clearly, the AIDS epidemic has changed forever the way FDA does its business. We do know that human subjects, patients, whether you consider infertility a disease or not, it is a treatment.

We recognize that risks are taken daily and that people want to take those risks. All we are asking, actually, is that when we don't know what the risks are, you should know that, too. If we have a means of addressing what the risk can be, we should, as a society, address that as well.

I don't think that, actually, the IND process is going to inhibit research at all. That was put to us for gene therapy for xenotransplantation. We think we have seen a lot more responsible and innovative research under regulation that is tailored to the risk and that we welcome everyone's participation.

Really, if we are wrong, you need to tell us that, as well. But, overall, thank you very much for coming to the table and saying that which you like and that

which you don't like. It is only by that way that we can do the right thing.

DR. SABLE: David Sable from St. Barnabas Medical Center in New Jersey. First, a note of appreciation for the quality of the meeting. It has really been outstanding.

Just two very minor factual points to clarify a couple of thing from the day. First, regarding cytoplasm transfer or ooplasm transfer, there seems to be a misperception that the procedure was designed to assist older women in conceiving. In actuality, it was designed to treat a very, very small percentage of women who we were seeing who seemed to have some defect in the ability of the eggs to function in the first two days after fertilization, mainly women in their mid to late thirties, whereas women in their forties and older seemed to be having problems more with the genetics of the nucleus.

It is a minor technical point but I wanted to clarify.

The other one, regarding preimplantation and genetic diagnosis, as far as I know, there are no programs that are blanket using it for any age group, that it is

still used in a case-by-case basis very specifically. We do it in approximately 14 percent of our cases and, in no case, do we look at any age group as being one that preimplantation genetics, as a blanket procedure, is used for.

DR. HUSZAR: I would like to ask a question from Alta. You mentioned about the regulation of the safety of the donor gametes. Among other things, I am the director of a sperm-donor program and we freeze our own sperm. So the interesting part of it is that, on the one hand, the studies show that when you are using donor sperm and you look at the recipient, they have just as high a percentage of abnormal karyotypes or all the different things that we can look at.

The second issue is that it is almost a bottomless well because we don't have all the tools. So every year, we will have new probes to look at more kinds of things we can test for. It really will make it extremely expensive if you have to do it.

Essentially, the realities that the normal-life people meet and they fell in love and they have a child, and they don't have any of that testing. So the question

is how much better we have to be than normal life and what is the definition of safety that we can live with.

MS. CHARO: I wish I could give you a concrete answer. Let me warn you that I cannot. We meet this situation frequently that we have two standards, one for personal activities and another for commercial or wide-scale activities that are aimed at marketing to the wider public. So, if I want to build a toaster to use in my kitchen, I am free to do so and it is very unlikely to meet all the usual product-safety standards.

But those standards kick in when I propose to somebody else that they use my toaster or my model of a toaster and I begin to market it widely. One of the reasons why a different standard exists in the latter is because of the concern that the usual sets of incentives and disincentives no longer apply.

If I am using something for myself and building it for myself, I have my own safety to be concerned about and it will probably deter me from being unduly sloppy about the electrical connections. But when we are in a more anonymous situation of purchaser and seller, some of

the those incentives to quality are missing and regulations pick up the slack.

There are other ways to create the incentives for quality, as I mentioned before, through things like the tort system or professional standards. So that is one reason, I think, why, in the formal donor programs, there has always been an expectation that the standard of safety would be higher than it is in ordinary intimate settings, although I do grant you that this is an initial question that is actually very difficult to discuss.

Whether that means that you have to use absolutely every possible means to eliminate every possible risk that could be known, I sincerely doubt because, in the area of medical practice, generally, we do not require optimization of the quality of medical services. We require a reasonable degree of care.

That term, however, is so elastic and so difficult to understand and so likely to bite you in the context of retrospective jury and judge reviews in the context of medical malpractice that what is considered reasonable care often has come to represent something

higher than the usual understanding of reasonable but still not 100 percent optimized.

I suggest that, in some ways, cost-benefit analysis will be a first cut. A second cut will be what does the typical consumer of these services expect. A third cut is going to be what do most other tissue banks and physicians do, and that the standard will lie somewhere in the midst of that kind of triumvirate of factors.

Each one of them, as they improve, will rachet up the standard for the other two. That is, professional standards get out in front with something written in a statement, it is going to ratchet up common practice, it is going to ratchet up consumer expectations and that, in turn, is going to change the cost-benefit analysis through economies of scale in some cases, et cetera.

So this is a moving target. I apologize that I can't be more specific.

MS. MADSEN: I actually want to respond to Philip. My concern over the IND process being introduced to our research is that infertility, unfortunately, is not covered in every state across this great land of ours. In

fact, there are more "have nots" when it comes to insurance coverage for infertility than "haves."

My concern is that infertility patients are now going to take on the costs of the IND process because I can assure you that the costs are going to be passed down to the consumer who is already very stressed out trying to pay for their treatment. So that is a piece of the concern from the patient perspective.

Another piece of the concern is I hear rumors, and, again, I am not a researcher so please correct me if I am wrong, that introducing the IND process to us, that within the IND process, things take much longer for research to actually be allowed to go to human trials and to reach us.

We have a very finite amount of time to build our families and there is a sense of urgency in our disease. My wish, and I know you have heard me say this before but now I am saying in front of the group, is, again, my wish is that this is what is going to be, that it needs to be streamlined and we need to be acknowledged to be a disease like cancer, that this process needs to be moved along and fast-tracked.

DR. LEPPERT: I am sorry, I came back here because I was up front and I didn't have a microphone. I am really very happy to have listened to everything that was said today and I am very pleased to hear people talking about the need for research. I do have one comment, however, and we tried to present this a little bit this morning, and that is that the Reproductive Sciences Branch does, in fact, fund a number of studies, albeit I know that it would be nice if we had more, but we have a number of clinical trials ongoing.

We have studies ongoing about endometriosis, polycystic ovarian disease. There are many things that are happening and we work with the FDA. In fact, in our clinical trials, we do get INDs for these clinical trials--it is funded by the NIH--so that people enrolling in the trials do not have to pay extra for being a clinical subject.

My question, I think, to this group is how do we work together as a community to get the research information out to clinicians and the public and also how does the Reproductive Science Branch let others know what we, in facta, can fund and have funded because even though

we have restrictions, we are able to do, within the framework of our mandates, a lot of things that really can help the whole question of what is healthy for mothers and fathers and children in the long term.

So my question to you is how can we further this dialogue and how can we help people know what research really is going on and what the results are.

MS. WARNER: Susan, could I just clarify one remark about the IND process because I think this is also important in the light of clarification that Phyllis was pointing out about the funding. But the IND model is actually a model for requiring research, clinical research, to follow certain standards of patient informed consent, subject informed consent, and also relying on some basis of data in order to form the basis for the investigation, to have a clinical plan, to have a protocol to follow it so that you are hoping to get useful results.

Another important part of the IND process is that it is not something that the researcher can charge for so the patients that are enrolled, it is experimental. It is understood as an experimental treatment and there is no

charge for that. We have a couple of exceptions, but that is the rule.

So, really, I guess what I would just challenge folks to take a look at is look at that as a model. Cancer, I think, has been mentioned as a disease. Cancer therapies are studied under the IND model. Basically, I think one of the advantages of that is that patients, in experimental situations, are not charged.

Then know it is experimental. They get the protections that are in place through the informed-consent process, through the IRB process, through FDA review and, ultimately, because of the results of those studies which are carefully designed, there is information upon which to base future choices about treatments.

The treatments that are successful come to light and those can be part of the myriad choices that are available for ART. The patient knows then, at that point, which ones work and which ones don't.

DR. WOOD: So now we are looking, again, for responses one to three.

DR. McLAREN: Just a word from the other side of the Atlantic. Anne McLaren. Mary Mahowald, I did enjoy

your little study about genetics versus gestation, the results of that. Nearly fifty years ago, I did a similar but much smaller study on women in England who I knew were reproductively active and the majority, but of course, it was not significant, came down on gestation rather than genetics.

But I didn't ask the men. And it never occurred to me. I wish I had.

But, more seriously, what I wanted to say arose both from what Mary Mahowald said and also from what Alta Charo said about the difference in ethical approach in different parts of the world because I think, in this country, USA, that the libertarian theory is really very dominant; reproductive freedom, freedom of research, freedom of practice of medicine.

In Asia, as Alta Charo mentioned, it is very much more the interest of the family, of the community, of the society rather than of the individual, themselves. I think Europe is probably somewhere in the middle but, as between United States and Europe, we believe in the same four basic ethical principles.

But I think, in Europe, we put more emphasis on justice and beneficence and in this country, I think there is more emphasis on autonomy and non-maleficence, if that is the right word. I think that probably has some effect on an appropriate regulatory system, but perhaps that can be explored more tomorrow morning.

MS. CHARO: I just want to respond to the point about the notion of justice because I think, actually, in my nonphilosopher's hat, that it is not actually so much a difference in whether or not there is an emphasis on justice. I think it is different notions of what justice constitutes.

It falls out, I think, not of the ethical analysis but of the constitutional differences between the member states of EU and the United States which very specifically has a constitution that starts with the premise of limited government and the protection of individual rights with a judicial gloss that says certain particular individual rights are given especially zealous protection from government infringement.

The ones listed in the Bill of Rights like speech and assembly are the ones most clearly understood to be

there. Marriage is another by virtue of court order, and reproduction in some of its aspects by virtue of court decision. And assisted reproductive technologies may be, maybe not, because we have never had a clear set of cases posing the question.

But there is something about some of the aspects of reproduction that is marked out for special protection. Now, why is this about justice? Because I think that the notion of justice that I have heard when I have attended meetings and participated in Europe on this topic is one that focuses on the greatest good for the greatest number, on social harmony.

In a sense, it is a kind of homogenous view of justice whereas, in the United States, this emphasis on individuals is not just about libertarian, do what you want, which is the old Marlboro Man image of the United States. It is about the idea that even an overwhelming consensus among most people is not sufficient to squelch the preferences of an eccentric and dissenting minority, that this would constitute an injustice that is greater than denying the majority their opportunity to create this harmonious society that I think, in Europe, people imagine

is more easily accomplished with laws that effectuate the popular will.

So I think it is a different notion of justice, justice in terms of greatest numbers versus justice not having any one segment of society particularly disadvantaged in order to let the rest of society proceed as they see fit that has caused such divergence. And it is why I have such skepticism about ever coming to harmonization in the way in which we regulate because I don't think it is so much a difference in the ethical analysis.

I think we all recognize these are close cases, whether it is England or the U.S. or France. We all wind up concluding they are close cases. I think what happens is that, in a close case, because of our different political and constitutional systems, we have different default positions which, in the United States, we default to individual preference and, in Europe, you might default to popular consensus, as a gross oversimplification.

DR. MAHOWALD: I agree entirely. I was attempting to contrast a libertarian conception of justice with what I would view as a more egalitarian conception.

Now, Amarcha Sen says everybody is egalitarian. It depends on equality of what that puts in one camp as opposed to several others.

I think, in some ways, in this country, this is a default position and I think that is problematic. I don't really believe that most bioethecists, these days, give the primacy to autonomy understood in a totally individualistic sense that, to some extent, the law is more likely to give as a default position.

But a position on justice which really does look at equitable distribution of benefits and harms among the populace is so much more complicated, an, I think, ideal, an ideal that may be better approximated but probably never achieved than a libertarian conception which simply says so long as we let and maximize the chance for all individuals to do what they wish, that the latter is pursued quite prevalently in our society.

My argument was that that is an inadequate position even in our own society because reproductive rights are not equally distributed in a free-enterprise system in which infertility treatment really is an industry.

I'll let it stop there.

DR. WOOD: We only have about five more minutes before we have to start moving out. I had pointed out two other responders.

DR. BAVISTER: Barry Bavister. I really wanted to make a comment about ooplasm transfer to Dr. Bonnicksen but I feel, in view of Dr. Leppert's comments, I would just like to take one minute to respond. Please don't think that my comments this morning were critical of Reproductive Sciences Branch. They were not intended to be.

But I just want to reiterate--you said how can you help. What we need to do is to change the way that grants are peer reviewed. They have to be peer reviewed, but, because the investigator-initiated RO1s, using appropriate animal models, are, by and large, being used as cannon fodder by CSR because of the triage system, you are not allowed to fund the grants you want to fund.

I see mission statements from Reproductive Sciences which are excellent, but I have tried to make the point before and failed that the NICHD cannot fund all the grants it would like to and is basically dictated to by CSR. That is the point I was trying to make. If that can

be changed, then a lot of things, a lot of the questions and problem brought up in this meeting, will be solved in good time by good research.

But right now, the research is doomed because we can't get the funding directed to the appropriate research studies. That is the point I am trying to make.

DR. SCHATTEN: Alta, you mentioned that, because your time was running short, you wouldn't speak about the issue of eggs as commodities. It strikes me that one of the other fanged creatures that is hanging over everyone's head, and I say this with affection to our FDA colleagues, is Congress in relation to the issue of cloning and the issue of embryonics, therapeutic cloning.

We are in a situation where eggs are, indeed, commodities and, until very recently, the only market really was infertility clinics primarily for the purposes of infertility treatment. Now, we have companies and other entities interested in buying eggs for the purposes of biotechnology. Can you elaborate on where you see the commodification of human oocytes going and what is a solution for that.

MS. CHARO: Actually, to be completely accurate, what I skipped over was disposition of embryos, not commodification of eggs. But you are right that that is one of the topics that caught everybody's attention last year. I think for some people it was a very sincere and legitimate concern and, for others, I think it became a rhetorical device by which they could sound more left-wing than they were as they pursued a right-wing agenda.

I think that there are many people who have engaged for years in a very basic debate that has not been resolved about whether all things should be available for commercial use or whether some things should be held off the commercial market for a variety of reasons having to do with sentiment and emotion.

This ranges from prostitution to--I actually prefer the term "contract motherhood," because I agree with Mary about the term surrogacy--to gametes to organs and non-organ tissue that is not reproductive, a whole variety of settings.

I believe that, to the extent that eggs are collected for reproductive purposes, they fall squarely within those debates because they are about the special

emotional content and, in the case where the egg is going to be used specifically to conceive a child and you are going to, therefore, have a biological connection to a child you don't rear, it raises those very issues about emotion.

I am a pragmatist so I don't believe it is possible to squelch markets completely. I think that, personally, a lot of what goes wrong in the management of things like prostitution and recreational drugs stems out of the fact that we try to keep it criminal as opposed to regulating it to death. So I probably would favor a regulatory approach even there as opposed to prohibitions.

I think with eggs collected for research purposes, you are in a different arena because it doesn't have the same emotional impact. Now it is really about body discomfort in exchange for payment. It is much more like going and doing uncomfortable or embarrassing circus work. That is a poor example. It is the end of the day. But you catch my drift.

It doesn't have the same kind of emotional significance as the reproductive use of one's gametes. So I would like to see them handled differently.

In either case, I think that both researchers and clinicians would find a wide variety of options that would probably keep everybody happier than they are now, in which payments were not excessive. Indeed, payments could be deferred. Give people savings bonds that don't mature for ten years. That is going to get rid of the quick kind of incentive for the college student who wants to spend it on beer or the person who has got a couple of kids who are going hungry and feels coerced to say, "Mine my ovaries."

There are a whole variety of things people haven't tried to entice without coercing in any sense of the word and allow for everybody's interest to be pursued.

DR. WOOD: As we approach the witching hour, are there any other urgent things you want to talk about today? I have one more over here.

DR. RACOWSKY: I would just like to respond to David Sable's comments regarding aneuploidy screening. David, in no way, shape or form did I mean to imply that your program was universally applying aneuploidy screening to a certain age group of patients. I just wish to say that. I am aware that at least one program, if now two, in our country are about to do, if not already doing,

aneuploidy screening in patients that are thirty-seven years or above in age uniformly across the board.

DR. SABLE: Thank you. I actually hadn't taken it personally.

DR. RACOWSKY: I'm glad. I thought we were buddies.

DR. SABLE: Thank you, Catherine.

DR. WOOD: Thank you, everyone. See you here at 8:00 a.m.

[Whereupon, at 5:45 p.m., the meeting was recessed to be resumed at 8:00 a.m., Thursday, September 19, 2002.]

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