Developing Cellular Therapies for Retinal Degenerative Diseases

Kapil Bharti,¹ Mahendra Rao,² Sara Chandros Hull,³ David Stroncek,⁴ Brian P. Brooks,⁵ Ellen Feigl,⁶ Jan C. van Meurs,⁷ Christene A. Huang,⁸ and Sheldon S. Miller⁹

¹Unit on Ocular and Stem Cell Translational Research, National Eye Institute, National Institutes of Health, Bethesda, Maryland

²National Institutes of Health Center for Regenerative Medicine, National Institutes of Health, Bethesda, Maryland

³Bioethics Core, Office of the Clinical Director, National Human Genome Research Institute, National Institutes of Health; and Department of Bioethics, Clinical Center, National Institutes of Health, Bethesda, Maryland

⁴Cell Processing Section, Department of Transfusion Medicine, Clinical Center, National Institutes of Health, Bethesda, Maryland

⁵Unit on Pediatric, Developmental and Genetic Ophthalmology, National Eye Institute, National Institutes of Health, Bethesda, Maryland

⁶California Institute for Regenerative Medicine, San Francisco, California

⁷Rotterdam University Eye Hospital, Rotterdam, Netherlands

⁸Department of Surgery, Massachusetts General Hospital, Harvard Medical School, Boston, Massachusetts

⁹Section of Epithelial and Retinal Physiology and Disease, National Eye Institute, National Institutes of Health, Bethesda, Maryland

Correspondence: Kapil Bharti, Unit on Ocular and Stem Cell Translational Research, National Eye Institute, National Institutes of Health, Building 10, Room 10B10, 10 Center Drive, Bethesda, MD; kapilbharti@nei.nih.gov.

Sheldon S. Miller, Section of Epithelial and Retinal Physiology and Disease, National Eye Institute, National Institutes of Health, Building 31, Room 6A22, 31 Center Drive, Bethesda, MD; millers@nei.nih.gov.

Submitted: October 19, 2013

Accepted: January 9, 2014

Citation: Bharti K, Rao M, Hull SC, et al. Developing cellular therapies for retinal degenerative diseases. *Invest Ophthalmol Vis Sci*. 2014;55:1191–1201. DOI:10.1167/iovs.13-13481

Biomedical advances in vision research have been greatly facilitated by the clinical accessibility of the visual system, its ease of experimental manipulation, and its ability to be functionally monitored in real time with noninvasive imaging techniques at the level of single cells and with quantitative end-point measures. A recent example is the development of stem cell-based therapies for degenerative eye diseases including AMD. Two phase I clinical trials using embryonic stem cell-derived RPE are already underway and several others using both pluripotent and multipotent adult stem cells are in earlier stages of development. These clinical trials will use a variety of cell types, including embryonic or induced pluripotent stem cell-derived RPE, bone marrow- or umbilical cord-derived mesenchymal stem cells, fetal neural or retinal progenitor cells, and adult RPE stem cells-derived RPE. Although quite distinct, these approaches, share common principles, concerns and issues across the clinical development pipeline. These considerations were a central part of the discussions at a recent National Eye Institute meeting on the development of cellular therapies for retinal degenerative disease. At this meeting, emphasis was placed on the general value of identifying and sharing information in the so-called “precompetitive space.” The utility of this behavior was described in terms of how it could allow us to remove road blocks in the clinical development pipeline, and more efficiently and economically move stem cell-based therapies for retinal degenerative diseases toward the clinic. Many of the ocular stem cell approaches we discuss are also being used more broadly, for nonocular conditions and therefore the model we develop here, using the precompetitive space, should benefit the entire scientific community.

Keywords: cell-based therapy, stem cells, age-related macular degeneration, retinitis pigmentosa

The National Eye Institute (NEI) in collaboration with the National Institutes of Health (NIH) Center for Regenerative Medicine (NIH CRM) organized a meeting to help advance and accelerate the field of stem cell-based therapies for retinal degenerative diseases. In these discussions, the NEI intramural program on induced pluripotent stem (iPS) cell research was used as a particular example, in order to be concrete and because it coincides with a primary outcome of the 2013 NEI Audacious Goals initiative to “Regenerate Neurons and Neural Connections in the Eye and Visual System” (<http://www.nei.nih.gov/audacious/>, in the public domain). Several stem cell-based therapies have been already proposed against degenerative eye diseases for the back of the eye. Some of the pioneering work in this field began from development of protocols to differentiate RPE from embryonic or iPS cells or

from adult RPE stem cells.^{1–11} Several different protocols are being developed for clinical-grade manufacturing. Researchers have found efficacy by using bone marrow- or umbilical cord-derived mesenchymal stem cells, and fetal neural or retinal progenitor cells in preclinical animal models.^{12–18} This meeting brought together a diverse group of international experts, associated with cell-based therapies from the public and private sectors (the Table includes a list of groups involved in developing cell-based therapies for the back of the eye), to advance and accelerate two main goals: (1) identify road blocks and find potential solutions for clinical application, production, and regulation of stem cell-based therapies for retinal degenerative diseases, and (2) promote collaborations among academic labs and private companies interested in these therapies.

TABLE. Clinical Studies Announced and Discussed at the June 24th to 25th Meeting

	Investigator/ Company	Cells	Trial	Protocol	Transplant	Clinical Stage	Disease
1	Advanced Cell Technology, Marlborough, MA	ESC-RPE	Allogeneic	Spontaneous differentiation	Cell suspension	Phase I/IIa ongoing	AMD-GA
2	Pete Coffey, UCL, London, UK, and Pfizer, Tadworth, Surrey, UK	ESC-RPE	Allogeneic	Spontaneous differentiation	Polyester scaffold	Trial begins early 2014	AMD-wet with rapid vision decline
3	PI - Mark Humayun, USC, Los Angeles, CA, Co-PIs - Dennis Clegg, USCB, Santa Barbara, CA, and David Hinton, USC, CIRM, San Francisco, CA	ESC-RPE	Allogeneic	Spontaneous differentiation	Paralene scaffold	IND filing 2014/15	AMD-GA
4	Eyal Banin, HMC, Jerusalem, Israel	ESC-RPE	Allogeneic	Developmentally guided	Cell suspension	GMP optimization, planning pre-clinical work	AMD-GA, Best disease, LCA
5	Masayo Takahashi, RIKEN, Wako-shi, Saitama, Japan, and Helios, Tokyo, Japan	iPSC-RPE	Autologous	Developmentally guided	RPE sheet, no scaffold	IRB approved, trial begins 2014/15	AMD-CNV
6	Pete Coffey, UCL and Pfizer	iPSC-RPE	Autologous	u.d.	Polyester scaffold	Planning	AMD-RPE tear
7	Kapil Bharti & Sheldon Miller, NEI, NIH, Bethesda, MD	iPSC-RPE	Autologous & allogeneic	Developmentally guided	Biodegradable scaffold	Planning cGMP optimization	AMD-GA, Stargardt's, RP
8	David Gamm, UW, Madison, WI	iPSC-retina/RPE	Autologous	Developmentally guided	Complex tissue	Planning proof-of-principle	AMD-GA
9	Sally Temple, NSCI, Rensselaer, NY	Adult RPE	Allogeneic	No differentiation	Cell suspension/ Scaffold	Planning cGMP optimization	AMD-GA
10	Janssen R&D, Beerse, Belgium	Umbilical tissue derived stem cells	Allogeneic	No differentiation	Cell suspension	Phase I/IIa ongoing	AMD-GA
11	Stem Cell, Inc., Newark, CA	Fetal neural stem cells	Allogeneic	No differentiation	Cell suspension	Phase I/IIa ongoing	AMD-GA
12	Henry Klassen, UCI, CIRM	Fetal retinal progenitors	Allogeneic	No differentiation	Cell suspension	GMP optimization, planning pre-clinical work	RP

CNV, choroidal neovascularization; ESC, embryonic stem cells; GMP, good manufacturing practice; IND, investigational new drug; iPSC, induced pluripotent stem cells; IRB, institutional review board. Clinical group affiliations: UCL, University College London; USC, University of Southern California; UCSB, University of California Santa Barbara; CIRM, California Institute for Regenerative Medicine; HMC, Hadassah Medical Center; NEI, National Eye Institute; NIH, National Institutes of Health; UW, University of Wisconsin; NSCI, Neural Stem Cell Institute; UCI, University of California Irvine.

In his opening remarks, NIH Director Francis Collins provided an elegantly clear and broad view of stem cell advances over the previous 10 years in the context of biomedical advances, challenges, and opportunities. He highlighted the particular advantages of the eye as an organ system for developing stem cell-based therapeutics, citing the recent striking breakthroughs in stem cell-based ocular organogenesis. More generally, he pointed out the extraordinary translational potential of iPSC cells and strongly emphasized the need for “disruptive innovation”¹⁹ as well as the importance of moving forward in collaboration with global collaborators. Both factors would be needed to overcome the scientific and technical barriers that lie ahead. He highlighted the unique set of characteristics and collaborative resources available in the intramural program including NIH Center for

Advancing Translational Sciences (NCATS) and the NIH Clinical Center. The former with high throughput screening to quickly analyze small molecule therapeutics, test for toxicity, and analyze pathways of disease, and the latter, the world's largest research hospital, to help facilitate and manufacture iPSC cell-based therapeutics, an immediate NIH goal.

Paul Sieving, the NEI Director pointed out the need for collaboration among groups working on the concept of cell-based therapies in the back of the eye (the Table includes a complete list of groups working on cell-based therapies in the back of the eye; also see Ramsden et al.⁷). The path to a clinical trial is long and the end points of successful clinical intervention are elusive, therefore requiring diverse collaborations to advance the field. He also pointed out that the eye is an ideal organ in which to begin trial therapies using stem cells

because of the optical and surgical accessibility of its internal structures and the broad and growing spectrum of noninvasive procedures that allow us to closely monitor clinical procedures. He presented the NEI Audacious Goals initiative “Regenerate Neurons and Neural Connections in the Eye and Visual System” to emphasize the overlap between the goals of this particular meeting to enhance and accelerate collaboration in the development of stem cell therapies against retinal degenerative disease and the just completed deliberations of a very large segment of the entire vision research community that found this to be a worthwhile target. He concluded with a quote from Einstein that “logic will get you from A to B, and imagination will take you everywhere.” The pioneering work completed by many dedicated people over the past several years provides a comprehensive portfolio that now allows us to consider and implement a wide range of clinical interventions using stem cell-based therapies.

The Director of the NIH Center for Regenerative Medicine, Mahendra Rao, began his presentation with a brief elaboration of the meeting agenda. He first pointed out the clear and urgent *a priori* need for stem cell-based clinical trial planning that includes tissue sourcing, clinical-grade manufacturing of cells, and preclinical animal studies. The present meeting was organized to induce discussions on all of these topics. He compared/contrasted the relative advantages and disadvantages of embryonic stem (ES) versus iPS cells as follows: (1) ES cells have been studied for a longer period of time as compared to iPS cells and are therefore better characterized, (2) ES cell-derivation methods allow the possibility of less genetic manipulation, (3) the biggest advantage that iPS cells provide is the possibility of autologous (and HLA-matched) transplants, which are immunologically more compatible with the host, (4) autologous transplants are likely to have fewer Food and Drug Administration (FDA) regulatory requirements as compared to allogeneic transplants, and (5) compared with ES cells, iPS cells are relatively easy to make, consent forms are less elaborate, and there are practically no ethical concerns with these cells. Economics is another important factor in considering stem cell trials based on the use of autologous versus allogeneic cells. Autologous cells can be more expensive and logistically challenging, but can be produced on a smaller scale using university/hospital scale good manufacturing practice (GMP) facilities.

A mandate of the NIH CRM is to facilitate stem cell-based clinical trials by developing and sharing resources in collaboration with the wider NIH community. For example, NIH CRM has developed contractual agreements with WiCell for clinical grade manufacturing of ES cells, and with Cellectics and Lonza for clinical grade iPS cell manufacturing. The NIH Clinical Center, Department of Transfusion Medicine (DTM) is currently developing capacity for the clinical manufacturing of iPS cell lines, tissue sourcing, cell storage, and the production of mature cells for transplantation. The NEI Intramural program is actively participating in these efforts with David Stroncek and his colleagues in the NIH Clinical Center Cell Processing Section. Both NEI and NIH CRM generated current Good Manufacturing Practice (cGMP) grade ES/iPS cells and NIH Drug Master Files (DMF) will be made available for cross-licensing and cross-referencing to other academic and private groups interested in similar efforts. The DMF is a document that contains complete information on all aspects of the cell manufacturing process and preclinical validation. Public-private partnerships provide intellectual synergy and cost reduction that benefit the biomedical community and ultimately, the American public.²⁰ Rao concluded by reminding us that it takes a village to plan and execute a clinical trial and it is a NIH responsibility to facilitate these collaborations.

Malcolm Moos, a senior investigator at the Center for Biologics Evaluation and Research (CBER)/Food and Drug Administration (FDA), provided the FDA perspective on stem cell-based therapies. He pointed out that for the approval of a phase I clinical trial, the main FDA concern is safety. More specifically, the mandate is that “human subjects are not exposed beyond reasonable and significant risk of illness or injury.” In addition to safety, FDA would also like to see data supporting product efficacy. He correctly pointed out that stem cells are a complex product, whose “critical quality attributes” are hard to define. Without the possibility of terminal sterilization, the product has inherent risks in terms of microbiological safety. Tumorigenicity and misdifferentiation are also potential concerns. It is often difficult to measure their efficacy, they have limited stability, a short half-life, and their heterogeneity biases cell sampling. Stem cells and their derivatives may include rare populations of cells that can provide beneficial or detrimental effects that may not be easy to quantify. His suggestion to the community is that we focus *a priori* on all these issues, perform better characterization and provide operational definitions of cell authenticity. Tighter specifications will improve product performance, reduce variability, and help us to meet endpoints goals during the pivotal efficacy trials.

Kapil Bharti, an NEI Earl Stadtman Investigator, summarized the ongoing NEI clinical efforts using iPS cells. He illustrated the value of collaborations and information sharing in the precompetitive space with a specific example involving autologous stem cell therapy. Traditionally, autologous stem cell therapy is usually given less consideration because it involves the use of a business model that is not economically viable. He suggested that the extra cost of this therapy can be mitigated by sharing DMF, clinical grade iPS cell lines, and other reagents needed for the investigational new drug (IND). For their iPS cell-derived RPE based clinical trial, the NEI team is planning to leverage several of the NIH intramural resources, including basic stem cell and RPE expertise, the clinical-grade manufacturing abilities of the Clinical Center Cell Processing Unit, the ability of the NEI clinic to manage a phase I/II clinical trial and long-term patient care, and the NEI/NIH ability to transfer technology/licenses to other centers. All of these features and the NIH commitment to provide training and expertise in various clinical aspects of stem cells will position NIH with characteristics described for an “Alpha Stem Cell Clinic,” a term recently proposed by the California Institute for Regenerative Medicine (CIRM, in the public domain at www.cirm.ca.gov) for a clinic capable of independently carrying out all the above mentioned tasks.²¹ To accelerate therapeutic development of stem cell therapies, CIRM has solicited proposals for creation of an Alpha Stem Cell Clinics Network, intended to provide an efficient, high quality, sustainable infrastructure to support clinical testing of investigational products, and eventual delivery of approved therapies, that will include consultative services, operational support, and a data and information management resource for informing the field and educating patients and the public. The NEI’s efforts in this direction involve the use of contracts made by NIH CRM to Cellectics and Lonza and the NIH Clinical Center cGMP facility. In order to optimize a cGMP-ready RPE differentiation protocol, Bharti has generated a TYROSINASE-GFP reporter iPS cell line. The generated iPS cell-derived RPE cells are grown on artificial biodegradable scaffolds to generate a polarized RPE tissue that has been functionally tested *in vitro*. He emphasized the need for an authenticated and physiologically relevant set of “release-criteria” for RPE cells and, as one example, suggested the measurement of apical to basolateral membrane fluid absorption across the monolayer to show that the cells are functional.²² He concluded with an offer of NEI

collaboration on different aspects of the clinical pipeline, including clinical grade iPS cell lines, reporter iPS cell lines, iPS cell to RPE differentiation protocol (research grade or clinical grade), transplantation tools, and DME.

The presentation by Bharti was expanded on by Alan Hubbs to provide details on technology transfer mechanisms and collaborative pathways at NEI. These opportunities were briefly summarized using examples from several NEI laboratories (Bharti, Miller, and Swaroop). He summarized and compared two different types of collaboration, one that used the NEI Material Transfer Agreement (MTA) with nonintellectual property (IP) terms. As a comparison, Hubbs also described collaborations that required a more formal cooperative research agreement where the IP terms need to be negotiated. Sury Vepa, a Senior Licensing and Patenting Manager at the NIH Office of Technology Transfer, described the path to licensing for several of the NEI generated technologies. National Institutes of Health policies allow for an exclusive license in the case of therapeutic drugs. Most technologies are licensed nonexclusively and know how is not licensed at all. Vepa also summarized specific examples for licensing technologies that were mentioned in Bharti's presentation.

CLINICAL TRIAL PLANNING

These discussions encompassed four main themes: Session 1 was on tissue sourcing; Session 2 covered GMP Manufacturing; Session 3 focused on Preclinical Animal Models; and Session 4 discussed From Animal Models to Clinical Trials.

Session 1: Tissue Sourcing

Sara Hull (see Appendix) began her presentation with the observation that "ethically weak biomaterial donation practices can undermine the research built upon them."²³ She further pointed out the mandate to respect the moral, religious, and ideologic views of volunteers who are the source of stem cells for all research and clinical applications. Commercial use of human biomaterial collected without appropriate consent can lead to both ethical and legal challenges. A notable example is the 21 human ES cell lines that were eligible for funding under the Bush administration. The consent forms associated with the use of these lines contained notable omissions and restrictions²⁴ that, for example, limited the ability to distribute them broadly and failed to inform participants about the risks of the loss of privacy. This had the effect of limiting the use of a number of these lines by the scientific community. A similar example, mentioned by Mahendra Rao, concerns the use of existing cord blood banks for making HLA-matched iPS cell line panels. These panels are NIH funded and the stored samples are HLA-typed. Unfortunately, most of those cord blood banks cannot be used because they lack the appropriate consent forms. These examples highlight the dual role of a robust informed consent that not only respects the study participants, but also protects the research outcomes and the precious resources that funded those research outcomes.

Two main types of consent models have been proposed and used in the past: the broad one-time consent, and the "sustained interaction" consent.^{25,26} The former model obtains participant consent solely at the time of specimen collection for unrestricted and unspecified future research use. Such consent has often been sufficient in the past for collecting patient samples for genetic analysis, but may not have informed participants about emerging issues such as public access to genetic databases, partnerships with pharma, and novel applications of regenerative medicine. The latter consent

model proposes a priori mechanisms for ongoing communication with donors to address some of these concerns and provides a means for re-consent as new potential uses and risks are identified. This model increases cost, donor and researcher burden, and it is logistically challenging to track "de-identified" stem cell lines to a particular donor. Currently, most biobanks are not set up to follow a "sustained interaction" model of obtaining consent.

Hull proposed an intermediate, hybrid consent model²⁷ that provides the possibility of a broad prospective consent for biospecimen collection that covers foreseeable research and for commercial applications such as disease analysis, large-scale screening applications, banking, distribution, and therapy. This model establishes boundaries around the uses of deriving reproductive tissue and the use of human cloning, and it describes potential reasons for recontacting the donor. Therefore, this consent form encourages "sustained interactions" to the extent feasible and with an option to opt out for the donor, within practical limits (item number 12 below). However, Hull acknowledged that "sustained interactions" are still a high-maintenance activity that will require a sustained costly infrastructure to maintain donor confidentiality.

Consent, as discussed above, relates to cell donors. Consideration should also be given to the issues surrounding informed consent for recipient participants in the clinical trials using stem cells. For both cell donors and recipients, the consent form should be explicit about what kind of information emerging from the research will or will not be returned to participants. Prior to submitting samples to cell repositories and permitting secondary use or distribution, the responsibilities of the investigators submitting samples, the repository and secondary users of samples should be clearly defined with respect to maintaining contact with donors, obtaining re-consent (if needed), limitations or restrictions on sample use, and returning results.

The guidelines for tissue sourcing and consent form considerations for stem cell-based clinical trials (ES or iPS or adult donor or fetal/adult cadaver stem cells) are as follows:

1. Potential risks and benefits of participating in a particular study should be clearly stated in the consent form;
2. The language of the consent form should be simple and easy to understand by each participant;
3. Appropriate language for all foreseeable intended uses of tissues, including broad research and potential commercial use of stem cells and their derivatives without any ownership claims by the donor or their next of kin should be included. This may include use as a therapeutic and/or a commercial product, ability to share stem cells, their derivative, and/or data obtained with them. The consent form should also state that donors may not have claims on patents issued on clinical grade stem cell lines generated from their material;
4. Adult donor stem cells and iPS cells are often obtained from a still living donor. Because the genetic information available through these cells can be potentially used to identify the donor, donor confidentiality needs to be protected by appropriate de-identification of the samples. This concern is exacerbated by the potential immortal nature of these cell types. If the samples are shared with other investigator, patient identifiers should not be shared;
5. The immortal nature of most stem cell types increases the potential to discover currently unforeseeable applications in the future for which specific consent was not obtained up front. Even with broad consent,

investigators should consider the possibility of re-consenting study participants for unanticipated uses, especially if new risks are identified;

6. The pluripotent nature of iPS and ES cells, and multipotent nature of adult stem cells also increases their applicability, translational, and therapeutic value. The consent form should be broad in terms of their use for different organs/tissues across the body;
7. Use of stem cells to derive reproductive tissue and use in human cloning goes beyond the general disease-related research uses that would be covered by a broad consent form. Any plans to use stem cell lines for these kinds of purposes must be mentioned explicitly in consent forms, separate from the broad consent described in number 3 above;
8. Permission should be obtained to perform whole-genome sequencing, genome editing, and animal (rodents, primates, etc.) transplantation experiments. Plans for managing the results of genome sequencing, including plans not to disclose results to the donors, should also be described in the consent form. For example, sequencing results obtained from stem cells may not be valid relative to the original patient material from which they were derived;
9. In the case of ES cells, refer to the detailed consent requirements for the donation of embryos. These are described as part of the NIH Guidelines for Human Stem Cell Research (in the public domain at <http://stemcells.nih.gov/policy/pages/2009guidelines.aspx>);
10. In the use of fetal/adult cadaver tissue, respect for next of kin views should be maintained while seeking permission for use in research/clinical applications;
11. Consent for an autologous therapy should be handled separately from the consent to donate specimens, with detailed descriptions of the transplant procedures and associated risks;
12. Limits to a donor's ability to withdraw from the study should be described up front. For example, donors can withdraw anytime and have their identifying information removed from samples and data, but under certain circumstances they will not be able to withdraw their cells or cell-derivatives. These include situations where cells have led to a research/clinical/commercial application that has been shared between labs or has high therapeutic potential;
13. In addition to the institutional review board (IRB) requirements for a consent form, researcher need to also follow FDA requirements for consenting the donors before tissue is sourced. The FDA (or an equivalent agency for other countries) donor eligibility Human Cell and Tissue Product (HCT/P) requirements for screening and testing should be met.

Session 2: GMP Manufacturing

A Good Manufacturing Practice (GMP) product is operationally defined by a set of "Critical Quality Attributes" that satisfy regulatory requirements. The first speaker in this session, Jiwen Zhang (see Appendix), pointed out that such Critical Quality Attributes (CQAs) can be achieved by a combination of correct tools and the appropriate collaborations between tool providers and cell therapy product developers. In other words, GMP is a systematic scientific approach that employs controlled production processes to manufacture FDA-regulated products. In the United States (US), FDA mandated GMP guidelines regulate manufacturing of potential drugs and devices for human use. The regulatory framework and defined

critical quality attributes for pharmaceuticals include safety, identity, purity, stability, and potency/biological activity of the product. The GMP requirements for cell-based products are the same as for small molecules and biologics, but are harder to define.

The GMP optimization of cell-based therapy products includes the scale-up of production from lab-grade and is often challenging since it limits product development and testing process. Most importantly, for stem cell-based products "the process is the product." Standard operating procedures (SOPs) need to be developed and validated for every step of the process. Depending upon the starting material (source/age/genetics/disease status), it is difficult to control process variability and its impact on critical quality attributes of the product. A significant challenge is that the final product is a live cell, it cannot be terminally sterilized. Therefore, raw materials and the process must be rigorously controlled. Another challenge presented by stem cell-derived products is that the relative viability of cells affects multiple critical quality attributes, including purity, potency, stability, and identity. This makes it hard to reproduce the process with defined SOPs. Some of these concerns can be addressed by shortening the culture duration and by developing closed and/or semi-automated culture systems. In summary, a continuous close collaboration between academic labs, product manufacturers, tool providers, and regulators is needed to develop regulatory standards for various cell therapy manufacturing platforms and will help achieve defined quality attributes for GMP development and production of stem cell-based therapies.

The International Organization for Standardization is commonly referred to as an ISO. This acronym refers to an international consortium to develop and publish SOPs and CQAs for goods and services of their member organizations. International Organization for Standardizations offer standardization procedures for both the public and private sectors and certification is a major step in allowing companies to confidently compete in a global economic community. For example, the WAVE Bioreactor, a single use closed bioreactor system with presterile disposable Cellbag, is an excellent example of an ISO certified product that meets all these GMP criteria. It is intended for early stage manufacturing of human cells in suspension and has been successfully used to manufacture clinical-grade T-cells and natural killer (NK) cells. The bioreactor is ISO certified for several biosafety attributes including in vitro cytotoxicity, local and acute systemic toxicity, cytotoxicity, irritation and sensitization, endotoxin, and sterility of cellular products manufactured in it.

These topics were taken up again in the context of an added complexity, global collaborations, that often include differing perspectives in the regulatory and manufacturing arenas. Toshio Miyata from the Japanese Ministry of Health, Labor, and Welfare (MHLW) introduced this discussion. Miyata, a cardiac surgeon by training, joined the MHLW with the goal of helping to reorganize policies for the accelerated approval of clinical trials in Japan. The MHLW is the main organization in Japan that approves GMP-based products. It seeks guidance from the Pharmaceuticals and Medical Devices Agency (PMDA), its technical arm that performs data analysis and scientific review for good lab practices (GLP) and GMP products in Japan. Two years ago the PMDA was reorganized and they opened a new Office of Cellular and Tissue-Based Products. In collaboration with institutions such as RIKEN in Kobe, PMDA has modified their guidelines for stem cell-based clinical trials and is now also taking a lead role in developing the guidelines for filing INDs. This has helped accelerate approvals for GMP products in Japan; for example, Miyata announced an autologous iPS cell-derived RPE clinical trial

led by Takahashi that is currently scheduled for initiation in 2014. These modified policies at MHLW should also allow for an earlier commercialization of regenerative medicine products. The MHLW now provides “adaptive licensing” for cell-based products with an earlier detection and evaluation of adverse events (safety) and probable benefits in a smaller patient population (e.g., 10–30 patients). This is followed either by marketing and a subsequent round of approval and evaluation on a larger patient population or by expiration of the initial approval. He also emphasized the need for simultaneous global development and highlighted the ongoing efforts between MHLW/PMDA and FDA to harmonize guidelines for cell-based product approval in Japan and the United States.

The presentations by Zhang and Miyata generated considerable discussion around two core topics enumerated below, the areas of collaboration and GMP requirements for cell-based INDs.

Some Critical Areas of Collaboration

Regarding biosafety standards, several international organizations (ISO, American Society for Testing and Materials [ASTM], Association for the Advancement of Medical Instrumentation [AAMI], and American National Standards Institute [ANSI]) have adopted standards for biosafety to ensure that contaminants can be avoided in the product, and there are no adverse effects of the manufacturing process on the product. Biosafety standards such as ISO 10993 that have been used in medical device and biologics GMP production are also applicable to cell or tissue therapy tools. Tool providers need to supply equipment and instrumentation that meet biosafety standards, but product manufacturers need to ensure proper utilization in the GMP process to obtain a microbiologically safe product. Such requirements are more stringent for allogeneic products that pose the risk of infectious agent propagation in a large population. Academic lab and product manufacturers have to work together to ensure that the product meets HCT/P FDA guidelines in section 21 CFR 1271.

When looking at performance standards, the performance of tools used in cell therapy manufacturing is crucial to achieve CQAs that are needed to ensure regulatory requirements for optimal product quality. Academic labs should invest time and efforts to carefully evaluate and validate tools' performance at an early product development stage to ensure successful production for the clinical study and commercial supply. Collaborations between academic lab, product manufacturer, and close interactions with regulators will ensure development of correct functional performance standards. Performance standards are in turn linked to cell characterization parameters. Cell characterization parameters include viability, identity, purity, and biological activity or potency. Organizations involved in developing cell characterization parameters and analytical tools to measure those parameters, include ASTM, National Institute of Standards and Technology (NIST), International Society for Cellular Therapy (ISCT), and the American Association of Blood Banks (AABB).

For documentation, tools, biosafety characterization, and performance validation need to be included in the Chemistry, Manufacturing, and Control (CMC) section of an IND. A commonly used mechanism to support an IND filing is to cross reference to a DMF. Reference to a DMF can aid the development of new cell-based INDs and facilitate collaborations between academic labs/private companies.

GMP Core Elements for Stem Cell-Based INDs. Labs and product manufacturers need to follow standard FDA guidelines recommended for cGMP manufacturing. Summa-

rized below are some specific concerns/challenges for stem cell-based products.

1. Facility and equipment: Stem cell-derived products are often cultured for long periods of time. Therefore, careful monitoring of aseptic working facility and equipment is needed. Disposable equipment and consumables should be used whenever possible. Closed systems provide an advantage to maintain aseptic working conditions. In the case where multiple autologous products are manufactured in parallel, dedicated equipment should be used for each product, if feasible, to avoid cross-contamination;
2. Personnel: Personnel need specific training to work with stem cells, for example, to derive/reprogram multipotent or pluripotent stem cell populations and to handle lineage specific differentiation protocols;
3. Supplies and reagents: All the materials used in the manufacturing process need to be tested in-house or a certificate of analysis (COA) needs to be obtained from the provider for the absence of adventitious agents. There are specific concerns with the use of xeno-material such as FBS, matrigel, feeders, and growth factors. Recommended guidelines are contained in 21 CFR 1271.290 (b);
4. Manufacturing process and record keeping: Manufacturing processes in the case of stem cell and their derivatives is also considered a part of the product. Standard operating procedures need to be established and validated for each step of the culture method. Critical manufacturing process controls should be established to monitor every production lot and for control of lot-to-lot variability. If needed, purification steps need to be developed and completely validated. Record keeping is specifically critical for stem cell-derived products because of long-term culture requirements. Autologous cell-based products need close monitoring and clear record keeping to avoid delivery of nonimmune compatible products;
5. Laboratory controls: For stem cell and their derivatives, major concerns are product consistency, purity, safety, stability, and potency. Therefore, lab controls should be established to quantify all of these critical quality attributes. For stem-cell derivatives, optimization of the manufacturing process is ensured in two ways: (1) the use of functional controls during manufacturing that analyze consistency, reproducibility, and efficacy of the process and therefore insure the release of significant quantities of therapeutic cell product, and (2) the use of controls that measure purity, microbiological safety, and potency of the product. Finally, a major challenge is the need for quantitative, reproducible functional assays that are monotonic with the potency of stem cell-derived products;
6. Packaging and distribution: Live stem cell-based products raise additional concerns for packaging and distribution. Utmost care needs to be taken to maintain aseptic packaging and distribution of stem cell-based products. Use of cryopreserved cells will aid in these processes.

Session 3: Preclinical Animal Models

The main theme of this discussion, led by Joy Cavagnaro (see Appendix), concerned the inadequacy, in many situations, of preclinical animal models of disease to fully or even partially model their human counterpart (see below).

Preclinical studies are intended to include: an established rationale for the clinical study and provide a mechanism of action; a rationale for dose escalation study starting with a safe minimum dose and therefore establish a pharmacologically effective dose for patients; an optimized administration route for cells/tissue transplant and show that the transplant can be safely delivered; and identification of potential toxicity from transplanted cells/tissues, characterize the toxic effect, and determine how often the toxic effect is seen.

Preclinical studies with cell-based products are fundamentally different than studies that use traditional small molecule drugs, mainly because stem cells and derivatives can survive in animals for much longer periods of time, can migrate to and integrate into any tissue, and can change cell fate/differentiation potential. Therefore, preclinical toxicology (including tumorigenicity) studies need to be performed for several months. There is a need to perform immunogenicity studies even if the cell-based product is autologous, but the need for these studies increases if the product is allogeneic. Efficacy studies need to be performed in a disease relevant model. Furthermore, the design of the preclinical study may change depending upon whether the product is a multipotent stem cell, a progenitor, or a stem cell-derivative and whether the cells are in suspension, encapsulated, or as sheets on a scaffold or without a scaffold. A preclinical study should be carefully planned because often efficacy data is interpreted over optimistically and toxicity data is dismissed as a species-specific difference. Safety endpoints are rarely included and a proper dose-response curve is often missing in preclinical studies. As Cavagnaro pointed out, preclinical studies need to satisfy FDA requirements and should be designed to provide answers for clinical trial decision-making.

Transplantation of human cells into an animal is by definition a xenograft study. It does not address the immunologic concerns of transplanting autologous or allogeneic human cells into human, but if designed carefully these preclinical studies can provide relevant safety and efficacy information about therapeutic products. Specific considerations for a preclinical study involving stem cell-based therapeutics in the back of the eye include the following:

1. Choice of animal model: It is important to perform preclinical studies in an animal model that can be informative for the clinical trial. Often more than one model might be required and this may depend on the kind of cell-based therapy (cell suspension or cell sheet) under evaluation. In the case of ocular cell-based therapies, both genetic and injury based models exist, and although the models do not fully represent the human disease they can be used to address specific questions about the cell-based product. For safety and biodistribution studies, small rodents might be sufficient. "Humanized rodent" models might be good choice for these kinds of studies.²⁸⁻³⁰ However, mice may not be appropriate to determine the efficacy of a RPE sheet. Mouse eyes are too small for transplantation of a cell sheet. The Royal College of Surgeon (RCS) rat model^{31,32} of retinal degeneration secondary to RPE dysfunction is currently a FDA-accepted model for efficacy studies involving stem cell-derived RPE transplantation. Rabbit, pig, or monkey models might be needed depending upon the size of the cell sheet to be tested. Devices used for surgery and the surgical procedure will also need to be tested in a large animal model with eyes comparable in size to the human eye. However, Huang pointed out that analogous cell types would need to be tested to avoid issues with receptor-ligand incompatibility between species, which would
- render results of human cell transplantation across most xenogeneic barriers uninterpretable and clinically irrelevant (e.g., CD172 and macrophage inhibitory receptor CD47 incompatibility)³³;
2. Study design and controls: Animal work should be designed to avoid surgery bias, for example: (1) if feasible, the use of product should be randomized against a placebo control in a masked fashion, (2) transplantation experiments can be staggered over several days/weeks, (3) different cell lots should be used, and (4) different individuals should perform surgery. Additional controls based on a particular model can, include sham surgery control, immunosuppression control, and transplantation device control. Assessment of the data should be masked. Adequate numbers of animals should be tested to ensure that the data are appropriately statistically powered; small rodents (10-20/sex/group/time point); nonrodents (4-6/sex/group/time point);
3. Dosing: Cell doses need to be chosen carefully to reflect the human situation. An appropriately scaled down, yet effective dose for small animals may not be easy to calculate. Dosing is more complex for a cell sheet; it may require different number of sheets or sheets of different shape/size. For the eye, particularly for macular degeneration, dose has been calculated according to the size of the macula (roughly 200,000 cells);
4. Efficacy endpoints: The eye allows easy noninvasive visualization of the transplant, which should be a standard routine test. In animal models, integration of the transplant can be easily demonstrated by histologic analysis. It is critical to use functional tests that provide an accurate indication of transplant function and visual function improvement. In the case of photoreceptor or RPE transplants, it may be necessary to test the function of both tissues. Behavioral and/or cortical recordings can be obtained to ensure proper visual recovery. Transplant survival should be monitored for several months and this should also guide the time course of efficacy evaluation;
5. Safety and toxicology endpoints: Delivery of cell suspension or cell sheet in the back of the eye is usually a minimally invasive procedure, but animals still need to be evaluated for damage caused by the delivery/surgery procedure per se (sham controls). Several aspects of this procedure can cause inflammation in transplanted animals; for example, surgery, cells, and the biomaterial for the scaffold. Antiinflammatory agents should be used after the surgical procedure and inflammation can and should be routinely monitored. Animals should be evaluated for inappropriate cell proliferation (tumors, ectopic tissue mass), ectopic differentiation, and migration to nontarget tissues. Food and Drug Administration guidelines require safety evaluations to be done for several months after transplantation. Spiking of transplants with undifferentiated ES/iPS cells can be used to test for purity and safety of a differentiated cell type. This provides data about minority populations of cells that may turn into tumors;
6. Toxicology studies, include changes in body weight, appetite, serum chemistry, blood cell count, blood coagulation, kidney and heart function, and the rate of mortality. Comprehensive animal pathology will need to be performed on most body organs. In some cases it may be useful to consider targeted studies in larger animal models (e.g., rabbits, pigs, or nonhuman primates) to confirm/optimize extrapolation of an efficacious dose as well as safety to humans.

Session 4: From Animal Models to Clinical Trials

Jan van Meurs (see Appendix), the leadoff speaker in this session, is a retinal surgeon whose work has formed the basis for stem cell-derived RPE transplantation in the back of the eye. He discussed his work on the autologous translocation of RPE in patients with exudative AMD. He pointed out that subsets of these patients that do not respond to anti-VEGF therapy, develop RPE tears, fibrotic scars, and hemorrhage, are likely good candidates for autologous RPE translocation.³⁴ In the initial attempts, this procedure was performed using a suspension of RPE cells isolated from the midperiphery of a patient's eye.³⁵ Injection of this RPE cell suspension under the macula did not result in any vision improvements, and often resulted in cell clumping and migration of some cells into the vitreous, leading to proliferative vitreoretinopathy (PVR). Bill Aylward isolated a healthy RPE sheet from an area adjacent to the macula and transplanted it to diseased macula.^{36,37} van Meurs later modified this procedure to obtain a full thickness RPE, Bruch's membrane, and choroid graft from the midperiphery allowing him to collect a larger sheet in a relatively more controlled fashion. This multilayered sheet was then transplanted in the affected macular area of the same eye. In all cases, the RPE graft was apparently revascularized from the remaining choroidal vessels.³⁸ In a unique example, a patient has been followed over 10 years with 20/32 best corrected visual acuity, reading vision, and little metamorphopsia (van Meurs J, personal communication, 2013). This approach has some limitations: (1) the graft itself is of the same age as the patient, and may reflect aging-associated changes, (2) it is often damaged by the surgical procedure, (3) the tissue tends to stick to the surgical device and has the tendency to curl at the edges, and (4) there may be intraoperative and postoperative hemorrhage complications leading to PVR. van Meurs concluded his talk by emphasizing that a transplanted autologous RPE sheet provides beneficial effects for a subset of AMD patients and this finding supports the concept of carrying out similar treatments using a stem cell-derived RPE sheet.

Christene Huang (see Appendix), the second speaker, highlighted the importance of transplant immunology for the success of a stem cell-based therapeutic.³⁹ She emphasized that the innate immune response, our first line of defense, is triggered by surgical damage, injury, infection, and by foreign material (e.g., scaffolds). Macrophages, NK cells, and granulocytes are the key mediators of this response. Innate immune activation, if left unchecked, can lead to activation of adaptive immune responses, including generation of cytotoxic T-cell (CTL) responses and antibody production against the foreign substance and against allogeneic cells. Therefore, one approach to diminishing the activation of an adaptive immune response is through control of the innate response following a transplant. For cell-based therapies, the source of cells can also have important immunological implications. The use of unmanipulated autologous cells would be the best option to avoid an immune response. Other sources that may invoke some degree of immune response include HLA mismatched allogeneic cells; HLA matched but minor HLA mismatched allogeneic cells; autologous cells with a xeno-antigen expression; autologous cells with an ectopic fetal antigen expression; and any xeno-material transplanted with cells (scaffold). Several strategies can be employed to combat antitransplant immune response in a host: (1) use of local and/or systemic immunosuppressive drugs, (2) enhancement of soluble immunosuppressive factors secreted by host cells or transplant itself may suppress the activation of responding cells, (3) tissues with low to absent expression of MHC class I may avoid damage from CTL responses, however, increased sensitivity to lysis by NK cells may occur,⁴⁰ and (4) immune privilege due to

preservation of intact barrier functions or local immunoregulatory properties of the host tissue; however, immune privilege may be compromised in a diseased state. Examples of immune modulatory mechanisms that play a role in controlling immune responses in the eye include the anterior chamber-associated immune deviation (ACAID) where exposure to allogeneic cells in the anterior chamber of the eye can induce regulatory T-cells that suppress T-cell-mediated immunity.^{41–43} More recently, it has been shown that RPE cells express the antiinflammatory TAM family of receptor tyrosine kinases, which play a role in controlling inflammation during 'homeostatic phagocytosis' of the outer segments of photoreceptors.^{44,45} Huang concluded by emphasizing that for a successful clinical intervention using a stem cell-based product, a combination of these approaches may have to be employed. She also emphasized that appropriate animal models are needed for proper assessment of immune responses to cellular therapy. The use of analogous cell types transplanted across allogeneic barriers would be much more appropriate than transplantation of human cells across xenogeneic barriers when assessing immunogenicity, cell survival, and even tumorigenicity.

Clinical Trial Design Considerations for Stem Cell-Based Therapeutics (Back of the Eye)

Regarding disease choice and patient selection criteria, the choice and stage of disease to target may depend on the mechanism of action of stem cell-derived products. There are two main categories, cells in suspension and cells in a sheet. In the case of stem cell-derived RPE, cells in a suspension probably function by providing nonpolarized trophic support or nonspecific phagocytosis. They may provide beneficial effects through the secretion of neuroprotective cytokines or neurotrophic factors and the ability to clear out debris in the host retinal space.^{46,47} If the cells do not integrate in the host retina/RPE/choroid layer, they may not survive over long periods. This approach may work in diseases where the area of atrophy is not extensive and the host tissue is able to respond to neuroprotective stimuli. Cells in a sheet provide polarized trophic support, specific receptor mediated phagocytosis, and vectorial fluid absorption from the apical to basal side. Of course, the cell sheet will have to integrate into the host layer for cells to function over longer periods. This is preferred in cases in which the area of atrophy is relatively large and a part of host RPE is nonfunctional.

Scaffolds are both biodegradable and nonbiodegradable. Biodegradable scaffolds provide the advantage that RPE cells can use the biological membrane that is laid out by them during in vitro growth and provides a platform for guided integration with the host RPE. However, because of their degradability they pose a significant regulatory challenge and may lead to short-term local inflammation. Nonbiodegradable scaffolds can more easily obtain FDA approval, but may not be as versatile inside the eye. For example, they may not allow transplanted RPE to integrate with the host RPE layer, and over longer periods may accumulate proteinaceous deposits that cause an inflammatory response and blockage of free nutrient/metabolite exchange.

Cell suspensions provide a huge advantage in delivery when considering surgery. Cells can be easily delivered via readily available, small diameter cannulas into the subretinal space, causing minimal retinal injury and forming a small bleb, which is quickly absorbed. In contrast, transplantation of a cell sheet requires a special delivery device that can hold and protect the sheet during the surgical procedure. The graft needs to be transplanted in the subretinal space in the correct apical-basal orientation, and a relatively large retinal opening is required, which may increase the risk of complications.

When controlling the immune response, local steroids will be required in every case to reduce surgery-associated inflammation. Allogeneic transplants may need long-term immunosuppression; currently both systemic and local immunosuppression have been planned. Systemic immunosuppression increases the risk of infections and tumor induction, whereas local steroid-based immunosuppression increases patient risk of developing glaucoma and cataract. Because inflammation can be easily monitored in the eye, careful monitoring of the patients will provide the possibility of modulating the immune suppression regime.

When considering the outcome variables, quantitative outcome measures should be carefully selected to follow the disease course and progression before transplantation to determine the clinical variability of a test, as well as the progression of a disease of interest. This will allow researchers to efficiently determine the efficacy of cell-based intervention. Although visual acuity is often a primary outcome variable in ocular therapeutic trials, it may not be ideal in all cases because of the state and nature of the condition; other functional and structural measures may also provide proof of efficacy, especially in early, proof of concept trials. One such example are studies, which showed that expansion of geographic atrophy in patients with the dry form of AMD directly correlates with improved visual acuity or function.^{48,49}

CONCLUSIONS

The notions of cooperation/connection are buried deep, some would say too deep, in the human psyche. However, the essays of Lewis Thomas posit that there is nothing “so communal, so interdependent” as science/biology (*The Lives of a Cell*, 1974; *The Fragile Species*, 1992).^{50,51}

The clinical potential of human embryonic stem cells was well appreciated following their first description in 1998,⁵² but progress toward translation was initially slow. In the past decade, scientific enthusiasm toward stem cells has significantly increased partly due to unprecedented technologic advancements, including the sequencing of the human genome, success with adult stem cell therapies, and most importantly, the discovery of human iPS cells in 2007.^{53,54} These scientific and technologic advances and the clear need for therapeutic interventions in a host of debilitating diseases, including retinal degenerative disease, have formed the basis for the clear anticipation of immense clinical progress in the near future. Stem cell-based therapy is currently considered as the “third pillar of medicine,” the first two being small molecules and biologics.⁵⁵

Stem cell-based therapies have complicated safety and regulatory requirements, because each therapeutic unit is a complex, highly interactive biological entity. The potentially wide-ranging interactive ability of these “units” greatly expands their therapeutic potential compared with biologics or small molecules. Currently, multiple therapies with different rationales and mechanistic approaches are moving forward to the clinic. Ideally, they can all move forward with the same vigor while eliminating overlapping sets of roadblocks. Although the current interest in clinical trials is high, it is critical to exercise extreme care and caution in these initial attempts to develop a successful cell-therapy for retinal degeneration. New field standards for safety and efficacy as well as IRB protocols and consent forms need to be jointly established in collaboration with the FDA and for use by the entire scientific community.

Based on historic precedence, the limitation in resources, and in the interest of time, we strongly believe that continued cooperation/collaborations are necessary for the development

of multiple successful clinical outcomes. It is anticipated that in the next 5 years several clinical studies will be completing their phase I safety trials. The success of these phase I trials depends on our ability to now cooperate in the solution of many as yet unanticipated regulatory and technical problems. We will then be at a critical crossroad given the much greater resources required for the implementation of phase II and III trials. Our ultimate goal is the widespread delivery of effective cell-based therapy for multiple retinal degenerative diseases. To achieve that goal, the success of current initiatives is critical and fundamentally dependent on our ability to work collaboratively across a broad based community.

The present discussion and previous considerations^{20,56,57} suggest an open-access model for accelerating the development of a cell-based IND for retinal degenerative diseases. Our open-access model is similar in approach to a recent public-private partnership initiative called Arch2POCM.^{56,57} This network of biomedical professionals (Arch) includes pharmaceutical companies, academics, and funding and regulatory agencies across the United States, European Union (EU), and Canada. Its goal is to exchange proof of clinical mechanism (POCM) for new potential drugs so that wider medical applications can be developed. This network would provide open public access to their phase II clinical data demonstrating safety and efficacy of a potential drug without any intellectual property claims. The hope is that negative POCM will help increase patient safety and reduce redundant efforts. We propose a similar active utilization of the precompetitive space to share information in the cell therapy field that would maximize patient safety and help shed light on redundant and apparently nonviable approaches. This goal suggests the following recommendations for investigators and companies in the four areas discussed above:

1. Develop consent forms that allow broad use of donor material and are accepted by multiple international regulatory agencies. These consent forms must also provide flexibility to accommodate radical scientific innovations and allow easy exchange of cell-therapy material across different trials. Both CIRM and the NIH CRM have developed and openly distributed consent forms that allow tissue sourcing for iPS cell based clinical trials;
2. Provide access (licensing) and cross reference to protocols, SOP, and DME. In a similar example, Cord Blood Banks openly shared their SOPs and manufacturing processes to help move the field forward (in the public domain at <https://web.emmes.com/study/cord/sop.htm>);
3. Develop and provide access to preclinical animal models, dosage, and toxicology studies performed with a given clinical-grade stem cell derivative. In a similar example, as part of an EU law, Registration, Evaluation, Authorization and Restriction of Chemicals (REACH), all EU companies producing chemicals are required to share their toxicology data in an effort to reduce redundant vertebrate exposure and minimize human risk (in the public domain at <http://echa.europa.eu>);
4. A “crowdsourcing”-like effort can be used to develop and share large-panels of clinical grade stem cell lines and their subclones (with reporter constructs) to accelerate preclinical safety and efficacy studies;
5. Develop transplantation and immune suppression procedures that are widely disseminated throughout the scientific community. Select and share disease outcome variables that provide accurate information about transplant success. As the number of transplant recipients grows over time, a shared database will allow the

possibility of generating statistically significant outcomes of cell therapy for rare diseases, ethnic backgrounds, tissue source of stem cells, age of recipient, stage of disease, and so on. A similar successful effort has been carried out for hematopoietic cell transplantation by the Center for International Blood & Marrow Transplant Research (in the public domain at <http://www.cibmtr.org/pages/index.aspx>).

To help transform these recommendations into specific guidelines, we propose an annual meeting of this group where experts from the academic sector, industry, funding and regulatory agencies, and patient-advocacy groups can all meet to determine an optimal set of specific guidelines that can be quickly brought to practice. We realize that the process of openly sharing intellectual property in a precompetitive space and the development of international databases for information sharing is a relatively new and daunting task. Nonetheless, we are encouraged and propelled along this shared path by strong synergies now developing for the creation of cell-based therapeutic interventions and for the analysis of disease. These efforts are energized and informed by public/private partnerships that extend over the entire biomedical community and include a strong public mandate.

Acknowledgments

The authors thank Jiwen Zhang (General Electric, Inc.), Joy Cavagnaro (Access BIO), Toshio Miyata (Ministry of Health, Japan), and Marco Zarbin (New Jersey Medical School) for helpful comments during the preparation of this document, and Cesar Perez-Gonzalez, Daniel Stimson, Dustin Hays, and Jennifer Wenger (National Eye Institute) for help in putting together meeting minutes.

Supported by the Division of Intramural Research, National Eye Institute, National Institutes of Health, and the NIH Center for Regenerative Medicine.

Disclosure: **K. Bharti**, None; **M. Rao**, None; **S.C. Hull**, None; **D. Stroncek**, None; **B.P. Brooks**, None; **E. Feigal**, None; **J.C. van Meurs**, None; **C.A. Huang**, None; **S.S. Miller**, None

References

- Buchholz DE, et al. Derivation of functional retinal pigmented epithelium from induced pluripotent stem cells. *Stem cells*. 2009;27:2427-2434.
- Carr AJ, et al. Protective effects of human iPS-derived retinal pigment epithelium cell transplantation in the retinal dystrophic rat. *PLoS One*. 2009;4:e8152.
- Klimanskaya I, et al. Derivation and comparative assessment of retinal pigment epithelium from human embryonic stem cells using transcriptomics. *Cloning Stem Cells*. 2004;6:217-245.
- Meyer JS, et al. Modeling early retinal development with human embryonic and induced pluripotent stem cells. *Proc Natl Acad Sci U S A*. 2009;106:16698-16703.
- Osakada F, Ikeda H, Sasai Y, Takahashi M. Stepwise differentiation of pluripotent stem cells into retinal cells. *Nat Protoc*. 2009;4:811-824.
- Osakada F, et al. In vitro differentiation of retinal cells from human pluripotent stem cells by small-molecule induction. *J Cell Sci*. 2009;122(pt 17):3169-3179.
- Ramsden CM, et al. Stem cells in retinal regeneration: past, present and future. *Development*. 2013;140:2576-2585.
- Reh TA, Lamba D, Gust J. Directing human embryonic stem cells to a retinal fate. *Methods Mol Biol*. 2010;636:139-153.
- Salero E, et al. Adult human RPE can be activated into a multipotent stem cell that produces mesenchymal derivatives. *Cell Stem Cell*. 2012;10:88-95.
- Schwartz SD, et al. Embryonic stem cell trials for macular degeneration: a preliminary report. *Lancet*. 2012;379:713-720.
- Vugler A, et al. Elucidating the phenomenon of HESC-derived RPE: anatomy of cell genesis, expansion and retinal transplantation. *Exp Neurol*. 2008;214:347-361.
- Aftab U, et al. Growth kinetics and transplantation of human retinal progenitor cells. *Exp Eye Res*. 2009;89:301-310.
- Inoue Y, et al. Subretinal transplantation of bone marrow mesenchymal stem cells delays retinal degeneration in the RCS rat model of retinal degeneration. *Exp Eye Res*. 2007;85:234-241.
- Lu B, et al. Human adult bone marrow-derived somatic cells rescue vision in a rodent model of retinal degeneration. *Exp Eye Res*. 2010;91:449-455.
- Lund RD, et al. Cells isolated from umbilical cord tissue rescue photoreceptors and visual functions in a rodent model of retinal disease. *Stem Cells*. 2007;25:602-611.
- McGill TJ, et al. Transplantation of human central nervous system stem cells - neuroprotection in retinal degeneration. *Eur J Neurosci*. 2012;35:468-477.
- Siqueira RC, Messias A, Voltarelli JC, Scott IU, Jorge R. Intravitreal injection of autologous bone marrow-derived mononuclear cells for hereditary retinal dystrophy: a phase I trial. *Retina*. 2011;31:1207-1214.
- Wang S, et al. Non-invasive stem cell therapy in a rat model for retinal degeneration and vascular pathology. *PLoS One*. 2010;5:e200.
- Kenagy JW, Christensen CM. Disruptive innovation: a new diagnosis for health care's "financial flu." *Healthc Financ Manage*. 2002;56:62-66.
- Rao M. Public private partnerships: a marriage of necessity. *Cell Stem Cell*. 2013;12:149-151.
- Trounson A, DeWitt ND, Feigal EG. The Alpha Stem Cell Clinic: a model for evaluating and delivering stem cell-based therapies. *Stem Cells Transl Med*. 2012;1:9-14.
- Li R, Maminishkis A, Banzon T, et al. IFN[gamma] regulates retinal pigment epithelial fluid transport. *Am J Physiol Cell Physiol*. 2009;297:C1452-C1465.
- Bahadur G, Morrison M, Machin L. Beyond the 'embryo question': human embryonic stem cell ethics in the context of biomaterial donation in the UK. *Reprod Biomed Online*. 2010;21:868-874.
- Streiffer R. Informed consent and federal funding for stem cell research. *Hastings Cent Rep*. 2008;38:40-47.
- Aalto-Setälä K, Conklin BR, Lo B. Obtaining consent for future research with induced pluripotent cells: opportunities and challenges. *PLoS Biol*. 2009;7:e42.
- Isasi R, Knoppers BM, Lomax G. Sustained interaction: the new normal for stem cell repositories? *Regen Med*. 2011;6783-792.
- Lowenthal J, Lipnick S, Rao M, Hull SC. Specimen collection for induced pluripotent stem cell research: harmonizing the approach to informed consent. *Stem Cells Transl Med*. 2012;1:409-421.
- Vatakis DN, et al. Using the BLT humanized mouse as a stem cell based gene therapy tumor model. *J Vis Exp*. 2012:e4181.
- Shultz LD, Brehm MA, Garcia-Martinez JV, Greiner DL. Humanized mice for immune system investigation: progress, promise and challenges. *Nat Rev Immunol*. 2012;12:786-798.
- Brehm MA, Jouvet N, Greiner DL, Shultz LD. Humanized mice for the study of infectious diseases. *Curr Opin Immunol*. 2013;25:428-435.
- Strauss O, Stumpff F, Mergler S, Wienrich M, Wiederholt M. The Royal College of Surgeons rat: an animal model for inherited retinal degeneration with a still unknown genetic defect. *Acta Anatomica*. 1998;162:101-111.

32. D'Cruz PM, et al. Mutation of the receptor tyrosine kinase gene Mertk in the retinal dystrophic RCS rat. *Hum Mol Genet.* 2000;9:645–651.
33. Navarro-Alvarez N, Yang YG. CD47: a new player in phagocytosis and xenograft rejection. *Cell Mol Immunol.* 2011;8:285–288.
34. van Zeeburg EJ, Maaijwee KJ, Missotten TO, Heimann H, van Meurs JC. A free retinal pigment epithelium-choroid graft in patients with exudative age-related macular degeneration: results up to 7 years. *Am J Ophthalmol.* 2012;153:120–127. e122.
35. van Meurs JC, et al. Autologous peripheral retinal pigment epithelium translocation in patients with subfoveal neovascular membranes. *Br J Ophthalmol.* 2004;88:110–113.
36. MacLaren RE, Aylward GW. Delayed atrophy of the retinal pigment epithelium after submacular surgery. *Euro J Ophthalmol.* 2005;15:170–172.
37. MacLaren RE, Bird AC, Sathia PJ, Aylward GW. Long-term results of submacular surgery combined with macular translocation of the retinal pigment epithelium in neovascular age-related macular degeneration. *Ophthalmology.* 2005;112:2081–2087.
38. van Zeeburg EJ, Cereda MG, van der Schoot J, Pertile G, van Meurs JC. Early perfusion of a free RPE-choroid graft in patients with exudative macular degeneration can be imaged with spectral domain-OCT. *Invest Ophthalmol Vis Sci.* 2011;52:5881–5886.
39. Loewendorf A, Csete M. Concise review: immunologic lessons from solid organ transplantation for stem cell-based therapies. *Stem Cells Transl Med.* 2013;2:136–142.
40. Long EO, Kim HS, Liu D, Peterson ME, Rajagopalan S. Controlling natural killer cell responses: integration of signals for activation and inhibition. *Annu Rev Immunol.* 2013;31:227–258.
41. Kaplan HJ, Streilein JW. Immune response to immunization via the anterior chamber of the eye. II. An analysis of F1 lymphocyte-induced immune deviation. *J Immunol.* 1978;120:689–693.
42. Kaplan HJ, Streilein JW. Immune response to immunization via the anterior chamber of the eye. I. F1 lymphocyte-induced immune deviation. *J Immunol.* 1977;118:809–814.
43. Stein-Streilein J. Mechanisms of immune privilege in the posterior eye. *Int Rev Immunol.* 2013;32:42–56.
44. Lemke G, Rothlin CV. Immunobiology of the TAM receptors. *Nat Rev Immunol.* 2008;8:327–336.
45. Prasad D, et al. TAM receptor function in the retinal pigment epithelium. *Mol Cell Neurosci.* 2006;33:96–108.
46. Bharti K, Miller SS, Arnheiter H. The new paradigm: retinal pigment epithelium cells generated from embryonic or induced pluripotent stem cells. *Pigment Cell Melanoma Res.* 2011;24:21–34.
47. Li R, Wen R, Banzon T, Maminishkis A, Miller SS. CNTF mediates neurotrophic factor secretion and fluid absorption in human retinal pigment epithelium. *PLoS One.* 2011;6:e23148.
48. Lee JY, Lee DH, Lee JY, Yoon YH. Correlation between subfoveal choroidal thickness and the severity or progression of nonexudative age-related macular degeneration. *Invest Ophthalmol Vis Sci.* 2013;54:7812–7818.
49. Querques L, Querques G, Forte R, Souied EH. Microperimetric correlations of autofluorescence and optical coherence tomography imaging in dry age-related macular degeneration. *Am J Ophthalmol.* 2012;153:1110–1115.
50. Thomas L. *The Lives Of A Cell; Notes Of A Biology Watcher.* New York, NY: Viking Press; 1974.
51. Thomas L. *The Fragile Species.* Cambridge, MA: Macmillan New Media; 1993.
52. Thomson JA, et al. Embryonic stem cell lines derived from human blastocysts. *Science.* 1998;282:1145–1147.
53. Takahashi K, et al. Induction of pluripotent stem cells from adult human fibroblasts by defined factors. *Cell.* 2007;131:861–872.
54. Yu J, et al. Induced pluripotent stem cell lines derived from human somatic cells. *Science.* 2007;318:1917–1920.
55. Fischbach MA, Bluestone JA, Lim WA. Cell-based therapeutics: the next pillar of medicine. *Sci Transl Med.* 2013;5:179ps177.
56. Norman T, Edwards A, Bountra C, Friend S. The precompetitive space: time to move the yardsticks. *Sci Transl Med.* 2011;3:76cm10.
57. Norman TC, Bountra C, Edwards AM, Yamamoto KR, Friend SH. Leveraging crowdsourcing to facilitate the discovery of new medicines. *Sci Transl Med.* 2011;3:88mr81.

APPENDIX

Session 1: Tissue Sourcing

Speaker: Sara Hull, Director Bioethics Core, Office of the Clinical Director, NHGRI, NIH; Chair: Barbara Karp, Combined Neuroscience Institutional Review Board Chair and Office of the Clinical Director, NINDS.

Session 2: GMP Manufacturing

Speakers: Jiwen Zhang, Regulatory Affairs Director, GE Healthcare; & Toshio Miyata, Ministry of Health, Labor, and Welfare, Japan; Chair: David Stroncek, Chief, Cell Processing Section, Department of Transfusion Medicine, Clinical Center, NIH.

Session 3: Preclinical Animal Models

Speaker: Joy Cavagnaro, Access BIO; Slides adapted from Joyce Frey-Vasconcells, Frey-Vasconcells Consulting, LLC; Chairs: Ellen Feigal, Acting President CIRM; Brian Brooks, Chief, Unit on Pediatric, Developmental and Genetic Ophthalmology, National Eye Institute, NIH.

Session 4: From Animal Models To Clinical Trials

Speaker: Jan van Meurs, Rotterdam Eye Hospital, Rotterdam, The Netherlands; Speaker: Christene A. Huang, Head, Hematopoietic Cell Transplantation Laboratory, Transplantation Biology Research Center, Massachusetts General Hospital, Boston, MA; Chairs: Eyal Banin, Director of the Center for Retinal and Macular Degenerations, Department of Ophthalmology, Hadassah-Hebrew University Medical Center, Israel.