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Review

Gene Therapy: Charting a Future Course—Summary of a National Institutes of Health Workshop, April 12, 2013

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Abstract

Recently, the gene therapy field has begun to experience clinical successes in a number of different diseases using various approaches and vectors. The workshop Gene Therapy: Charting a Future Course, sponsored by the National Institutes of Health (NIH) Office of Biotechnology Activities, brought together early and mid-career researchers to discuss the key scientific challenges and opportunities, ethical and communication issues, and NIH and foundation resources available to facilitate further clinical advances.

Introduction

Recently, after some years of deferred hopes, a number of gene therapy trials have demonstrated clinical benefit, including trials for congenital eye diseases (Hauswirth *et al.*, 2008; Maguire *et al.*, 2008), hereditary immune system disorders (Aiuti *et al.*, 2009; Hacein-Bey-Abina *et al.*, 2010; Booth *et al.*, 2011), adoptive transfer of genetically engineered T cells for cancer (Frantz, 2011; Porter *et al.*, 2011; Grupp *et al.*, 2013), and gene therapy for hemophilia B (Nathwani *et al.*, 2011; VandenDriessche and Chuah, 2012). In 2012, a gene therapy approach for lipoprotein lipase (LPL) deficiency (Stroes *et al.*, 2008) became the first gene therapy product licensed in Europe. As the field

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is poised to build on these initial successes, this is an opportune time to consider the scientific and technological challenges, ethical and communication issues, and the resources and tools needed to facilitate efficient translation of gene therapy into clinical practice.

To explore these issues, a workshop titled Gene Therapy: Charting a Future Course was organized and sponsored by the National Institutes of Health (NIH) Office of Biotechnology Activities (OBA), Office of Science Policy, Office of the Director. The overall goal was to gather early and mid-career researchers in conversation with senior scholars in the field to discuss the key scientific and technical advancements; critical resources that are needed; and the emerging ethical, biosafety, and communication issues that might be expected to arise as the field expands. The workshop was attended by investigators from academia, representatives from NIH and foundations that support gene therapy research, corporate stakeholders, staff from the Federal Food and Drug Administration, and members of the NIH Recombinant DNA Advisory Committee (RAC). The following is a summary of the discussion and represents the views of the individual authors, not those of NIH. The webcast and slide presentations from this workshop are available for review on the OBA website.

Scientific Frontiers for Gene Therapy

Currently, there are over 1200 gene therapy trials registered with OBA. While the first licensed gene therapy product in Europe is for a rare disease, and there have been a number of successful trials for rare inherited immunodeficiencies and eye disorders, the majority of gene therapy trials target cancer. This is a maturing field and licensed products in the United States are also expected in the near future. However, the majority of protocols registered with OBA from 2010 through 2013 are for early phase trials: 75% are phase I or phase I/II trials, 21% are phase II, and 4% are phase III. Approximately one-third of the protocols registered during this period proposed to use retroviruses (including lentiviruses), followed by DNA plasmids (~19%), adenoviral (Ad) vectors (~15%), and adeno-associated virus (AAV) vectors (~8%).

To review the current state of the science and define some of the technical and other challenges, the meeting began with presentations of novel approaches to the design of vectors or cell-based systems, and current and upcoming clinical applications. In the field of oncology a number of different approaches are being pursued, including immunotherapy, oncogene downregulation, tumor suppressors, vector-directed cell lysis, and suicide genes. In addition, there have been a number of successful trials for rare diseases, in particular ocular and inherited immunodeficiency disorders, as well as other inherited neurological and muscle disorders.

Oncologic applications

Cancer immunotherapy—in particular, T-cell immunotherapy—is a major area of research as discussed at a recent NIH workshop, which highlighted the breadth of research taking place in this arena (http://osp.od.nih.gov/office-biotechnology-activities/event/2013-09-10-123000-2013-09-11-161500/scientific-symposium-t-cell-immunotherapy-optimizing-trial-design). Dr. Nabil Ahmed, Baylor College of Medicine, summarized the research on genetically engineered T cells for cancer immunotherapy and highlighted some of the work being done at his institution. The field has pursued two general approaches: (1) genetically modified T-cell receptors (TCRs) or (2) chimeric antigen receptors (CARs), which are artificial receptors, the prototype design of which includes a variable domain of a tumor-specific antibody combined with a T-cell receptor signaling domain (Eshhar *et al.*, 1993; Pule *et al.*, 2003). While there have been a number of trials showing promising clinical responses, key challenges and questions that remain include how to enhance T-cell survival and expansion (Pule *et al.*, 2008; Savoldo *et al.*, 2011), whether targeting of multiple antigens will be required to increase efficacy (Grada *et al.*, 2013; Hegde *et al.*, 2013), and whether the inclusion of suicide genes (DiStasi *et al.*, 2011) will ensure efficient and timely elimination of genetically engineered T-cell products. Future directions will be aimed at the development of off-the-shelf and broad-spectrum products that can be rapidly produced (Gerdemann *et al.*, 2013).

Rather than being a final treatment after standard therapy, the promise of cancer immunotherapy may only be realized when it is integrated with other therapies such as surgical debulking, chemotherapy, or other immunomodulating agents (Vanneman and Dranoff, 2012). Dr. Daniel Sterman, University of Pennsylvania, described his experience studying different gene transfer approaches for pleural mesothelioma, a disease particularly suited for gene therapy because the tumor is relatively localized and can be repeatedly accessed to evaluate tumor and immune responses. Initial studies were conducted with Ad vectors expressing the suicide transgene, herpes simplex virus thymidine kinase (HSV-TK). More recent studies focused on Ad vector delivery of interferon transgenes to the pleural space with the goal of eliciting a global immune response that includes the generation of antitumor cytotoxic T lymphocytes. Strategies to improve immunotherapy approaches include treating earlier stage research participants and defining and modifying their specific immunosuppressive tumor microenvironment.

Ophthalmologic applications

The eye has proven to be an excellent early target for gene therapy, offering the ability to directly transduce the defective cells in a relatively immune-privileged site for viral vector readministration. Dr. Shannon Boye, University of Florida, described efforts to extend the success of AAV gene therapy for *RPE65*− Leber congenital amaurosis (LCA2) to other LCA genotypes. LCA1, one of the most common forms of LCA, is caused by a defect in the guanylate cyclase 1 (*GUCY2D*) gene that encodes GC1, a protein expressed in photoreceptors. Patients with LCA1 have no cone function; however, their retinas are preserved and present a target for gene therapy (Jacobson *et al.*, 2013). AAV vectors derived from various serotypes with different promoters drove GC1 expression in photoreceptors, restored retinal function and visually guided behavior, and preserved retinal structure in multiple animal models of GC1 deficiency (Boye *et al.*, 2010, 2011, 2013). On the basis of its ability to drive transgene expression exclusively in photoreceptors (foveal and parafoveal cones and rods) of nonhuman primate, a clinical

trial is being planned involving subretinal injection of AAV5-hGRK1-*GUCY2D* (Boye *et al.*, 2012).

Dr. Boye also discussed two challenges faced in ocular gene therapy—how to avoid surgical trauma when delivering vector to patients with inherited retinal degeneration, and how to overcome the limited carrying capacity of AAV in order to treat diseases associated with mutations in large genes. Ocular gene therapy to date has focused on subretinal delivery, which is a relatively complex surgical procedure. Dr. Boye discussed how to safely deliver therapeutic genes to photoreceptors in fragile, degenerate retinas via intravitreal delivery with vectors that can transverse the inner limiting membrane to reach photoreceptors. To accomplish this goal, an understanding of AAV receptor biology and intracellular trafficking is required. Using rational design and directed evolution, AAV capsids are being evaluated for transduction efficiency after intravitreal injection in a mouse model containing sortable photoreceptors. To overcome the limited carrying capacity of AAV (~5 kb) to allow for the delivery of larger genes for other retinal disorders, several dual AAV vector platforms are being investigated. Halves of large genes can be coadministered by two AAV vectors. Those halves can then recombine to form a full-length gene once inside the target cell. Dr. Boye demonstrated the feasibility of using such dual AAV vector platforms to reconstitute *Myosin7A*, a large gene associated with Usher syndrome 1B (Lopes *et al.*, 2013; Dyka *et al.*, 2014).

Dr. Luk H. Vandenberghe, Harvard Medical School, discussed the possibility of extending gene therapy to other ocular diseases, such as LCA caused by other mutations or other retinal disorders caused by single-gene mutations, through the development of a platform based on the commonalities in the diseases and similar AAV vectors. However, because the ocular field has a wide spectrum of monogenetic disorders, it is first important to conduct natural history studies, which will be essential for the evaluation of efficacy, determination of biomarkers and end points, and selection of subject population. He acknowledged that there are also opportunities to improve vector design, not only by developing standardized pharmacological assays, and a more clinically practical method for intravitreal injection, but also by methods for regulating transgene expression.

Gene transfer for muscle disorders

Another long-standing area of interest for gene therapy trials is the muscular dystrophies. While there are a number of protocols focused on gene replacement, Dr. Scott Harper's laboratory at Ohio State University College of Medicine focuses on the use of RNA interference (RNAi) to treat dominant myopathies by silencing mutant alleles. Dr. Harper described his studies of limb girdle muscular dystrophy 1A (LGMD1A), a myopathy in which mutations in the myotilin (*MYOT*) gene cause the formation of MYOT protein aggregates that disrupt the myofibrillar apparatus. In an LGMD1A mouse model, *MYOT*-targeted miRNAs knocked down MYOT expression, reduced aggregate formation, and improved muscle mass and strength (Liu *et al.*, 2014). While these results suggested that RNAi should be a useful approach for dominant negative disorders, Dr. Harper noted a number of hurdles to translation, including immunology issues, need for natural history studies, and the challenges of navigating from preclinical studies through preparation of investigational new drug (IND) submissions to the FDA and large-scale vector production.

Advances in ex vivo *approaches using hematopoietic stem cells and induced pluripotent stem cells*

The earliest gene therapy trials focused on combined immune deficiencies, using correction of gene defects in hematopoietic stem cells (HSCs), and there have been notable clinical successes reported in multiple trials for adenosine deaminase deficiency–severe combined immunodeficiency (ADA-SCID) (Candotti *et al.*, 2012) and for X-linked severe combined immunodeficiency (Hacein-Bey-Abina *et al.*, 2010), although for the latter this clinical success came with an unacceptable rate of vector-driven hematologic malignancy. Dr. Satiro De Oliveira, University of California–Los Angeles reviewed the uses of $CD34^+$ HSCs transduced *ex vivo* with vectors to deliver transgenes for immunodeficiencies (Aiuti *et al.*, 2009; Hacein-Bey-Abina *et al.*, 2010; Booth *et al.*, 2011), sickle cell disease (Romero *et al.*, 2013), and β thalassemia, and the potential to use such cells in cancer immunotherapy (e.g., for gene modified T-cell receptors, or CARs) (De Oliveira *et al.*, 2013; Giannoni *et al.*, 2013). Challenges for the next generation of clinical trials include development of efficient, minimally genotoxic vectors, standard toxicology studies, large-scale production of clinical-grade vectors, and multiphase, milestone-driven research support.

Expanding beyond HSCs as gene therapy targets, Dr. Yasuhiro Ikeda described his work at the Mayo Clinic with induced pluripotent stem cell (iPSC) approaches for diabetes. In their lab, biopsy samples from different somatic tissue types were transduced with lentiviral vectors expressing the four genes necessary for reprograming to iPSCs. These patient-derived iPSCs were then guided to differentiate into insulin-producing cells, however, with significant intrapatient variation in differentiation (Thatava *et al.*, 2013). In a mouse model, transplantation of these cells led to teratoma formation, possibly because of insertional mutagenesis caused by the lentiviral vector or long-term expression of the reprogramming factors, which include the *c-myc* oncogene. The use of nonintegrating Sendai virus vectors to decrease the risk of teratomas is currently being tested (Kudva *et al.*, 2012). While this is a promising approach, the complex nature of iPSC products raises many challenges to be overcome before clinical use, including the need for good manufacturing practice (GMP)–grade reprogramming vectors, reagents used in differentiation, long processing time, possible autoimmunity issues, and favorable safety records to balance risk and benefit in a diabetic population with other therapeutic options.

Improving gene delivery vectors

Many of the ophthalmologic protocols, as well as protocols for hemophilia, muscular dystrophy, and pediatric neurological diseases, have used AAV vectors. While there have been a number of successes, immune responses and gene transfer to off-target tissues are challenges that could limit this vector's ability to treat a number of diseases. To build a stable source of AAV vectors, Dr. Aravind Asokan at the University of North Carolina is investigating how to

re-engineer AAV strains to decrease off-target transduction, increase transduction efficiency in order to decrease needed vector doses, and escape preexisting immunity (Asokan *et al.*, 2012). Many AAV serotypes have been isolated that bind to different receptors, thus displaying different tropisms, which could be useful for specific clinical applications. In order to fine-tune viral vector tropism, Dr. Asokan used the knowledge of the various receptor footprints to manipulate the molecular determinants of tropism by modifying amino acid side chains to develop, for example, AAV9 vectors that were detargeted from the liver in order to increase transduction of heart and skeletal muscles (Asokan and Samulski, 2013), as well as AAV4 vectors with improved CNS tropism by modifying the interaction with mucin. A similar approach is being used to develop vectors that can evade preexisting immunity by manipulating the footprints of antibody–antigen interactions. The field of AAV vector development has expanded from the few serotypes available in the 1990s, to the discovery of many more natural serotypes, to now using a rational engineering approach to design vectors for specific applications. Dr. Asokan pointed out that a better understanding of cross-species variability in AAV tropism and transduction efficiency among different preclinical animal models is critical for guiding future trials and possibly predicting clinical outcomes. Further, it is clear from ongoing studies that despite their diverse tissue tropisms, the safety profiles of various AAV strains have been largely similar, thereby emphasizing the importance of standardized protocols, safety assessment, and analysis of AAV vectors.

The successes, failures, challenges, and opportunities in the future for gene therapy as a drug delivery platform were summarized by Dr. Ronald Crystal, Weill Cornell Medical College. Compared with other drug delivery systems, gene therapy has advantages such as sustained and local delivery, steady-state levels, delivery of intracellular proteins, and the ability to introduce novel functions into cell types. The field has succeeded in developing a number of well-defined vector systems designed for different target cells resulting in clinical benefits in several indications, including LPL deficiency, immunodeficiencies, hemophilia B, retinitis pigmentosa, and leukemia. Much is still unknown about these systems however. Among the lessons learned from previous clinical studies are that innate and adaptive immunity may be an obstacle; intravascular administration has dose limits; not all cells can be modified in an organ; and it is more difficult to downregulate than to upregulate genes. He noted that a key to the successes seen to date is the ability to target clinical diseases that have a measurable phenotype.

In addition, he noted that there are a number of unmet needs in the field, such as the need to improve vector safety and efficacy. He urged the field to establish standards for vector production and focus on developing controlled gene expression. The field should consider developing vectors that can be targeted into specific sites in the genome or that carry genomic DNA, rather than focusing on cDNA transgenes.

Finally, as the field matures, it is hoped that it will move toward developing treatments for more common, acquired disorders and milder disorders. In addition, it may expand into complex etiologies, such as underlying biological mechanisms of psychiatric disorders (e.g., addiction). The ability to develop safe vectors and have established data to support clinical safety will be critical. Obviously, as the field moves from incurable malignancies or fatal rare diseases to approaches to treat more common and more complex conditions for which there are effective medical or surgical interventions, the risk–benefit analysis will shift. However, he argued that the limited funding resources available for academic research may limit both study size and choice of indication, so partnership with pharmaceutical, biotechnology, or venture capital companies will likely become more critical if less public resources are available.

Ethical Considerations and Public Perceptions

There are significant bioethical dimensions to all basic and clinical research, not only to gene therapy. What may be unique to gene therapy is it comes out of a history of significant skepticism and even fear about the biologic, social, and ethical consequences of genetic manipulation. In the 1970s, questions regarding the safety of genetic manipulation in nonclinical research and fears about unintended consequences led to public demand for oversight. Rather than be subject to regulation, the field agreed to self-regulation and a local oversight system was developed governed by federal guidelines, the *NIH Guidelines for Research Involving Recombinant or Synthetic Nucleic Acid Molecules* (http://osp.od.nih.gov/office-biotechnology-activities/biosafety/nih-guidelines). At the national level, this system was characterized by transparency and public engagement, with the inclusion of nonscientists, such as lawyers and bioethicists in decision making. In the more than 40 years since the advent of recombinant DNA technology, the demonstrated benefits to basic research and the development of various pharmaceuticals, as well as the increased understanding of the associated risks, have led to greater acceptance of this technology for many applications.

Clinical research with recombinant DNA started in the early 1990s and the oversight process, with its transparency, reassured the American public as this technology moved into human applications. Gene therapy applications being targeted today are confined to somatic cell manipulation; however, for some people not directly involved in its application, the perception remains that gene therapy is targeting the fundamental essence of life largely because of the increasing popular association of personal identity with nuclear DNA. In addition, because gene therapy had a challenging and vexed early history, with promise dramatically overstated and few tangible rewards at hand, there was significant public concern when a clinical gene therapy trial led directly to the tragic death of a young man, Mr. Jesse Gelsinger (Zallin, 2000). This tragedy fundamentally changed the field and the public perceptions of its usefulness and of the ability of government oversight to adequately monitor the complexities of a clinical trial, especially those in which significant financial conflicts of interest were involved and large amounts of money were at stake.

This history surely supports the need for rigorous and impartial oversight on such issues as informed consent and refusal processes, transparent data safety and monitoring systems, conflicts of interests, and ethical issues. This is especially true as first-in-human trials are brought forth based on animal data and basic science theories and as the field moves forward toward new therapies and cures. Researchers must be conscious of not overpromising, remaining

transparent, and being honest about their successes and failures. This history also underscores the need for engaging the public because the new science is advancing rapidly, and the public may contribute important new questions about the nature, goal, and meaning of the work.

Finally, gene therapy's history, with the initial promise and excitement giving way to a greater understanding of the complexities of the science after a tragic death, may still drive the perception of the field. Some individuals, in both academic and lay circles, may still see gene therapy through lens colored by its problematic history as an overpromised technology. Dr. Laurie Zoloth pointed out that in a recent *New York Times* article about positive results for hemophilia gene therapy, the author noted that the disease appears ''treatable by gene therapy, a technique with a 20-year record of almost unbroken failure'' (Wade, 2011). While those in the field would not agree with this assessment, critics in other disciplines understand this history as a widespread cautionary tale about science and its promises. It may take a far stronger and more widespread series of successful trials to strengthen the acceptance of gene therapy as an established medical therapy.

This history may also influence the tolerance for risk from gene therapy. Whether this perceived lack of tolerance for risk in new gene therapy trials reflects a general shift regarding risk acceptance in any medical research or something specific to gene therapy is unclear, with some arguing that gene therapy has been unfairly constrained because of its history and others providing counter examples (Deakin *et al.*, 2009; Kimmelman, 2009).

Resources: What Is Available and What Is Needed?

Obtaining funding for basic and clinical research is a priority for all of the researchers. The participants noted the need for researchers and institutions to leverage funding resources, in addition to NIH grants, for clinical trials. Foundations are one such source available to investigators. Foundations represented at the meeting included the Foundation Fighting Blindness, National Tay-Sachs & Allied Diseases Association, Inc., Muscular Dystrophy Association, Alliance for Cancer Gene Therapy, and Batten Disease Support and Research Association (Table 1). Similar to NIH grants, their process includes scientific peer review scoring; however, their focus is on developing new innovative therapies for their specific diseases. These foundations have successfully provided significant translational funding for clinical work.

The NIH supports a variety of programs that facilitate translational research. The National Heart, Lung, and Blood Institute (NHLBI) offers support for translational gene therapy research through its Gene Therapy Resource Program (GTRP). The GTRP offers preclinical vectors and immunology testing; GLP-compliant, IND-enabling pharmacology and toxicology studies; GMP-grade AAV and lentivirus vectors; and partial clinical trial funding assistance and regulatory affairs support to qualified investigators. These services are provided at no cost to the investigator.

Another NHLBI-supported resource is the National Gene Vector Biorepository (NGVB) and Coordinating Center, located within the Department of Medical and Molecular Genetics and the Indiana University School of Medicine. The NGVB houses a repository that includes a wide range of plasmids, cell lines, and other reagents submitted by gene therapy investigators to facilitate broader access to these novel agents. The NGVB offers archiving services for specimens from clinical trials and pharmacology/toxicology studies, as well as for reserve or back-up clinical-grade vector and master cell banks. The NGVB also maintains a

Table 1. Resources Available for Gene Therapy Research

NIH resources
Gene Therapy Resource Program (GTRP) (www.gtrp.org)
National Heart, Lung, and Blood Institute (NHLBI)–supported program designed to facilitate translation of gene transfer applications into clinical interventions
Core facilities for preclinical and clinical good manufacturing practice–grade vectors (adeno-associated virus, adeno, and lentivirus), pharmacology, toxicology, and immunology studies, and a clinical coordinating center
Clinical trial performance and regulatory application support
Genetic Modification Clinical Research Information System (GeMCRIS) (www.gemcris.od.nih.gov/Contents/GC_HOME.asp)
Information resource and analytical tool that allows public users to access basic reports about gene transfer trials and develop specific queries (e.g., vector, transgene, medical condition)
Rare Diseases Clinical Research Network (RDCRN) (http://rarediseases.info.nih.gov/ASP/resources/extr_res.asp)
Program coordinated by the Office of Rare Diseases Research, National Center for Advancing Translational Science (NCATS), to support and facilitate research by creating consortia of research for related rare diseases with the goals of cost sharing, establishing uniform protocols, and collaborative clinical research
Clinical and Translational Science Award (CTSA) (www.ncats.nih.gov/research/cts/ctsa/about/about.html)
Program within the Division of Clinical Innovation, NCATS, provides funding to individual universities to increase the efficiency and speed of clinical and translational research across the country.
Foundations Funding Gene Transfer Research
Foundation Fighting Blindness (www.blindness.org/)
National Tay-Sachs & Allied Diseases Association, Inc. (www.ntsad.org/)
Muscular Dystrophy Association (http://mda.org/)
Batten Disease Support and Research Association (www.bdsra.org/)
Alliance for Cancer Gene Therapy (www.acgtfoundation.org/)

database of pharmacology/toxicology studies previously conducted to support FDA applications for gene therapy clinical trials. Upon request from an investigator, the NGVB will assist with securing letters of cross-reference to allow the FDA to use this information when reviewing an investigator's IND application. The NGVB also offers testing by linear amplification-mediated PCR and ligation-mediated PCR technologies for clonal expansion of cells on specimens from clinical trials for which the FDA requires post-trial monitoring of participants.

NIH recently formed the National Center for Advancing Translational Science (NCATS). An important program for academic researchers is the Clinical and Translational Science Awards (CTSA). The mission of this program is to catalyze the generation of innovative methods and technologies and enhance the development, testing, and implementation of diagnostics and therapeutics. Nationwide, there are currently 62 sites at academic health centers that have NIH-funded CTSAs. The overarching goals of each site and of the national program are to facilitate training and education of investigators in the full spectrum of translational science bridging between the bench, the clinic, and the community and promoting improvements in the quality, safety, efficiency, and cost-effectiveness of clinical and translational research (Davis, 2012). Investigators can access CTSAs at their institutions. Gene transfer research has been supported by CTSA at the University of California–Los Angeles (De Oliveira *et al.*, 2013) and University of Washington (Till *et al.*, 2013) for CAR T-cell research.

Many gene therapy protocols focus on rare diseases, and clinical trials for rare diseases raise unique issues, as the number of subjects available for trials may be limited. In addition, good data on the natural history of the disease are often lacking and this may make it more difficult to develop efficacy endpoints. In response to some of these challenges, in 2003, the NIH Office of Rare Diseases (ORDR), which is now part of NCATs, created a Rare Diseases Clinical Research Network program. The program was developed by ORDR in collaboration with a number of NIH institutes. The network has created research consortia that focus on multiple related diseases. Each consortium must have two multisite clinical studies, one of which must be a longitudinal study, such as a natural history study or genotypic–phenotypic correlation study. The consortia involve multiple investigators and sites and collaborate with patient advocacy groups and foundations. The goal is to facilitate clinical research by making large-scale clinical studies possible through the development of uniform protocols for data collection and cost-sharing infrastructure. All consortia are linked by a single data management and coordinating center that provides technologies and tools to collect standardized clinical research data and can also provide support for study design and data analysis. In addition, the program seeks to ensure that there is collaboration with patient advocacy groups, and there is training for new investigators. Currently, there are 17 consortia with 2290 members, including 97 patient advocacy groups and foundations. There are 86 clinical protocols that are accruing research participants. In addition, 174 fellows and new investigators have been trained in clinical research on rare diseases.

With regard to needed resources, participants noted that the field would benefit from funding mechanisms that extend from vector development and preclinical testing through the initial phases of clinical trials. Once in the clinic and beyond the initial phase I or small phase II studies, a potential hurdle will be the ability to scale-up production. Currently, vector production is decentralized across a number of institutions. This may be challenging to continue as products move to commercialization, but as some products rely on gene modification of autologous cells, this may remain a model for certain applications. There was general agreement that industry support would be needed for optimization of production for commercial application.

Regulatory Frameworks and Commercial Development

One issue raised by investigators was the need for consistency in review standards. The invited mid-career investigators were largely from academia, with their clinical studies being funded by NIH or other grants. Many of their protocols are subject to review by the funding organization, the NIH Recombinant DNA Advisory Committee, the FDA, and locally by the Institutional Biosafety Committee and Institutional Review Board. It was understood that these bodies have different mandates and expertise. Also noted was the possibility for inconsistency within a review agency or body, if over time the staff at an agency or a review committee changes. A new reviewer or committee member could raise new issues not raised by the previous reviewer. Because grant funding is done in 4–5-year cycles, responding to new recommendations can sometimes make it difficult to reach the grant endpoint in the time allotted.

Another problem highlighted was the intellectual property (IP) issues that can arise with these products. In the manufacturing of new gene therapy agents, the vectors, transgenes, promoters, and reagents used may be covered by different IP claims. IP issues may present a barrier to collaboration across institutions for academic researchers. Some investigators have found that the multiple material transfer agreements (MTA) that must be signed in order to access reagents for product development are sometime onerous. One investigator wondered whether NIH could require that any product developed with NIH funding uses a universal MTA that standardizes this process. For pharmaceutical companies, the costs associated with obtaining multiple licenses for components of a potential gene therapy product may make it less attractive to pursue such a product compared with one that has fewer IP constraints. While the meeting participants agree with the need to protect IP, navigating the licenses for reagents and component may eventually impede progress. The field may need to consider innovative strategies to help overcome some of these hurdles.

Finally, participants voiced uncertainty regarding how gene therapy products would be reimbursed by insurers and how that might impact the willingness of pharmaceutical companies to invest in bringing promising products to market. Many drugs involve chronic administration resulting in a revenue stream over time. A number of gene therapy applications seek to provide long-term therapeutic benefit with only a single application, much in the way bone marrow transplant does. However, bone marrow transplant is not a commercial product. If there is uncertainty regarding the ability to recoup investments from these therapies, it may be difficult to attract the investment needed to bring these products forward.

Biosafety and Long-Term Follow-Up

Gene therapy is unusual among therapeutics in the use of modified viruses and bacteria that can potentially spread into the environment and because a number of vectors have the potential to persist for months or even years. In all clinical protocols, there is an assessment of immediate safety issues, but gene therapy has also required assessment of long-term safety. Currently, the vast majority of gene therapy trials have enrolled subjects with metastatic or advanced malignancies. While there have been some notable successes and long-term survivors, on the whole, the focus on end-stage malignancies has limited the available long-term follow-up data. In the past few years, long-term data on children treated with retroviral vectors in trials for several primary immunodeficiency disorders have been published (Fischer *et al.*, 2013; Mukerjee and Thrasher, 2013). In addition, several other publications on long-term follow-up after Ad vector administration for cardiac disease (Kaminsky *et al.*, 2013) and AAV vectors for Canavan's disease (Sondhi *et al.*, 2012) have also been published. Many additional protocols are beginning to collect long-term data; however, not all of these data will be published. Meeting participants agreed that a central repository for such data that could be accessed by investigators and the public would be an important resource that could be used to support new trials, could allow safety data to be monitored and assessed, and could lead to the eventual licensing of gene therapy products. As OBA has access to much of these data, a central repository could possibly be created by a natural extension of the current GeMCRIS database.

Another potential issue is the biosafety implications of certain viral vectors, in particular replication-competent and oncolytic vectors. While vectors such as AAV are not associated with human disease, shedding of a modified herpes or vaccinia virus may raise public health concerns. Even for vectors that are not likely to lead to human disease (e.g., a nonreplicating vector derived from an attenuated virus), it may be important for public acceptance to be able to

Table 2. Opportunities and Challenges Ahead and Resources Needed

Current challenges	*Future efforts*
Need for improved understanding of the role of specific disease factors in gene therapy outcomes	Natural history studies to help with • identification of disease targets with measurable phenotypes • selection of subject population • identification of biomarkers • determination of end points • evaluation of efficacy
Ability to understand and manage immune responses to vectors	Vectors designed to evade preexisting immunity Immune-suppression approaches
Minimizing the risk of toxicity, including genotoxicity from integrating vectors that are used for long-term gene correction	Development of appropriate and validated preclinical assays to help predict clinical experience • determine whether data from previous studies could be usefully shared (e.g., biodistribution of sufficiently equivalent vectors)
	Modifications to vectors to decrease the risk of insertional mutagenesis, e.g., • integration into safe harbors • integration-deficient lentiviral vectors • genome editing
Controlled transgene expression	Development of vectors with • controlled gene expression • genomic rather than cDNA transgenes
Refining tissue targeting and improving transduction efficiency	Vectors targeted to desired cell type Determination of dose limitations for different routes of administration
Data sharing	Enhancement of a central repository accessible to investigators and public • preclinical data for possible cross-referencing for IND applications (e.g., expansion of NGVB pharm/tox database) • long-term follow-up study data (e.g., GeMCRIS)
	More frequent publication of this type of data • e.g., *Molecular Therapy Methods, Human Gene Therapy Methods, Human Gene Therapy Clinical Development*
Resources for translational research and early clinical trials	Multiphase, milestone-driven research support Industry involvement
Commercialization	Large-scale production of clinical-grade vectors Establishment of reference standards Standardize MTA across NIH-funded institutions Pricing models for one-time clinical therapies Reimbursement models that recognize single-dosing therapies and will facilitate access

IND, investigational new drug; MTA, material transfer agreements; NGVB, National Gene Vector Biorepository.

reference expert guidance to confirm that there is a low level of risk and explain why it is appropriate to deliver such vectors in an outpatient setting. For replication-competent viruses, it is critical to generate guidance on whether initial isolation is warranted and what type of monitoring for shedding or other precautions and counseling should be done. Every trial with such potential should include a plan for this risk. Ideally, these plans would be generated by investigators, informed by guidance established by experts in the field.

Conclusions

The NIH OBA organized this workshop to consider the most important scientific opportunities on the horizon and to identify the challenges to realizing these opportunities, including knowledge gaps, policy issues, ethical concerns, and needed resources. The introductory talks touched on a few of the many promising areas of gene therapy, and new applications and successes continue to be seen. For example, in the T-cell immunotherapy area, several reports on gene therapy trials received much scientific and media attention when significant clinical responses were seen in adults and children with advanced leukemia who had exhausted other approved therapies (Grupp *et al.*, 2013; Davila *et al.*, 2014). These exciting results highlight the enormous promise of cancer-targeted T cells. Also in the past year, the OBA has received a number of protocols proposing first-in-human approaches for rare diseases that build on the recent successes in this area.

Yet even as this field begins to see efficacy, scientific challenges remain. Participants highlighted the need for continued refinement of vectors to reduce the risks of immune reactions and allow for better organ or cell targeting, for example, intravitreal delivery for retinal diseases. If vector production methods could become more uniform across protocols, this would facilitate the comparison of results across clinical trials and could enable investigators to build more efficiently on these experiences. Further work should be done to determine how best to integrate gene therapy approaches into other modalities of care, in particular for cancer, as the combination of chemotherapy and gene therapy may provide the optimum benefit.

Together with the need for early career support, the nature and timeframe for that support are critical issues. For investigators new to the field, early funding that enables them to focus on their research rather than needing to balance other demands, such as clinical duties, may be the key to launching a gene transfer research program successfully. For clinical applications, the ideal funding would extend from proof of concept, through preclinical work, vector production, and the initial clinical trial. In addition to adequate funding, other resources considered important included developing uniform and streamlined MTA for reagents and other materials necessary for gene therapy products developed with the support of NIH funding. In the regulatory arena, investigators stated that greater consistency across regulatory reviews, both among institutions and within agencies, should be a goal.

While some of these issues are not confined to gene transfer, it was also recognized that these products may raise specific issues that need to be addressed by the community. The potential public health risks from replication-competent vectors derived from viruses associated with human disease may be low, but nonetheless promoting consistent guidelines for the management of research participants who have received such vectors will be a priority, and OBA will seek to facilitate this process with investigators. In addition, as many gene transfer products are able to persist for a number of years, a means for sharing long-term safety data across clinical trials would benefit the field, and is another area that would benefit from NIH stewardship.

The discussions at this workshop helped assess the scientific opportunities and challenges ahead, and resources needed to assist the field in its progress (Table 2). As the field approaches what may be its next phase, entering mainstream medical practice, this group of mid-career investigators offered their ideas for funding and policy initiatives that may facilitate this transition.

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