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# TRANSLATING GENE THERAPY TO THE CLINIC

## TECHNIQUES AND APPROACHES

EDITED BY:
JEFFREY LAURENCE AND MICHAEL FRANKLIN

# Translating Gene Therapy to the Clinic

## Techniques and Approaches

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# Preface

In the summer of 1968 two staff scientists at Oak Ridge National Laboratory, Stanfield Rogers and Peter Pfuderer, suggested in a letter to *Nature* that "viral RNA or DNA information" could be used in the control of genetic deficiency diseases as well as nonheritable disorders such as cancer.[1] Their proposition was based, like many scientific breakthroughs, on an experiment of nature: the observation that circulating arginine levels are elevated in humans following infection with Shope papilloma virus, which was thought to induce a virus-specific arginase. Theirs was a prescient thought, borne on the eve of the birth of recombinant DNA technology. But it took some four decades to begin realizing its promise.

The first approved use of gene therapy occurred in 1991 under the direction of W. French Anderson. Ashanti DeSilva was a four year old girl with the enzyme-based immune deficiency disorder ADA-SCID. Retrovirus-mediated transfer of an adenosine deaminase gene into her autologous T cells led to a clinical response, albeit incomplete and temporary.[2] This was followed by a "loss of innocence" attendant on the treatment, in 1999, of Jesse Gelsinger, an 18 year old man with ornithine transcarbamylase deficiency.[3] He died as a consequence of an adenovirus vector-associated inflammatory process. Shortly there after five infants with SCID-X1 developed acute leukemia after receiving a murine retrovirus-based gene therapy to replace a defective interleukin 2 receptor H chain.[3] But then only one of those five patients died from their leukemia. And with a final enrollment of 20 SCID-X1 infants, and correction of severe immune deficiency in 17 of them over a median follow up of 9 years, gene therapy was finally established as a realistic therapeutic for those without alternatives.[4]

Since that time over 1800 gene therapy trials in 31 countries have been initiated or completed.[4] And the field's promise is not restricted to "simple" replacement or excision of a defective gene. For example, genetic engineering techniques have been used to inculcate tumor recognition or virus resistance in autologous lymphocytes of patients with metastatic cancer[5] and advanced AIDS.[6] Although there are currently no U.S. FDA licensed gene therapy products, in 2012 Glybera (alipogene tiparvovec) became the first example of this technology to be approved for clinical use, in Europe, after its endorsement by the European Medicines Agency.[7] Based on an adeno-associated virus type 2 (AAV2) vector, Glybera corrected a defect in the lipoprotein lipase gene that otherwise leads to severe pancreatitis. Like most new technologies Glybera is expensive—about $1.6 million per patient—partially related to the ultra-orphan nature of the target disorder. (There are only a few hundred cases annually in the resource rich world.[7,8]) But its clinical success, as well as preliminary data from phase 1/2 and phase 3 clinical trials for more common conditions, as outlined in our text, has led to an explosion in commercial interest; between 2013 and early 2014 US companies have invested some $600 million in gene therapy research.[9]

The text you are about to explore is an introduction for the translational and basic researcher as well as the clinician to the vast field of gene therapy technology. It is the first book in a new series, *Advances in Translational Medicine*. The project is a direct outgrowth of our editing of an illustrious journal, *Translational Research, The Journal of Laboratory and Clinical Medicine*. It is coincident with the journal's celebration of a legacy of 100 years in the promotion of excellence in clinical and translational research. This first volume is also a perfect opportunity to congratulate the Central Society for Clinical and Translational Research (CSCTR), a key partner with the journal. Albeit technically only in its 87th year, CSCTR traces its heritage to the Central Interurban Clinical Club, the establishment of which, in 1919, places it not far off the 100-year mark. Its fundamental goals, shared by our journal and this series, are critical and constant. Above all, champion the young investigator, bring in new ideas, establish diverse collaborations, and limit inbreeding. The special topics issues published annually in *Translational Research* are highly quoted. They achieved sufficient notice that the book division of Elsevier, publisher of our journal, began this series based upon expanded versions of our special issues and invited reviews.

Early on, the national importance of our society was well recognized. It also had an unanticipated impact on gene therapy related issues. The policies of genetic modification in clinical trials are regulated by the Declaration of Helsinki. And in 1966, only four societies were requested to endorse this declaration relating to "ethical principles for medical research involving human subjects"; the American Medical Association, the American Society for Clinical Investigation, and the American Federation for Clinical Research joined us. This declaration, along with the 2001 HUGO (Human Genome

Organization) consensus, covers the types of somatic gene therapies discussed in our text. Germline gene approaches by which gametes (sperm or ova) are modified, permitting a therapeutic manipulation to be passed on to future generations, are proscribed for ethical reasons in many countries, and are not covered here.

The authors of the following chapters are leaders in the field of gene therapy. They cover a range of topics and technologies with a depth and clarity to be commended, providing helpful illustrations and comprehensive citations to the literature. Several chapters focus on specific diseases, while others cover new technologies or barriers to progress. It strives to cover, in depth, disease-specific areas of particular promise. Its initial focus is on mechanisms of introducing a gene, generally via a viral vector, that either: (1) causes a protein to be expressed in a patient with a defective protein product, or two little of the normal one; or (2) introduces editing genes, "molecular scissors" that excise or disrupt genes causing a disease. As the field has evolved to encompass non-DNA-based technologies, utilizing antisense oligonucleotides, small interfering RNAs, etc. that do not alter a gene, but directly interact with RNAs or proteins, are also presented here.

These chapters also provide roadmaps to the ontogeny of current gene therapy trials and methods by which a group might design their own. I have borrowed a recently published patient-centered approach to designing a gene therapy for epilepsy[10] as an example of how the introductory chapters of this text set the stage for strategies to tackle your own areas of therapeutic interest.

1. Choose an animal model that accurately reflects the clinical problem in which to conduct preclinical studies.
2. Decide on a therapeutic approach. This is simpler when a single-gene defect is involved, limiting a functional protein product correctable by a relatively small increase in that product, as in hemophilia B. In a complex phenomenon such as epilepsy, one needs to decide if the target might best be decreasing neuronal excitation or increasing neuronal inhibitory pathways. Targeting of an entire cohort of genes could be contemplated.[10]
3. Choose a safe, effective vector. At the moment this usually means AAV, in which case limited payload size is a major impediment, or a lentivirus. But retrovirus, adenovirus, herpes simplex virus, plasmid, and other transport systems are also in various stages of clinical testing, and are outlined herein.
4. Consider all potential obstacles and explore them. Our text considers issues of payload toxicity, vector targeting, sufficiency of gene product expression, and the limits of in vitro and animal models. It also touches upon potential regulatory issues and good manufacturing-practice costs, but related details are left to other sources. For example, the American Society of Gene & Cell Therapy offers Web sites with information on issues related to the conduct of clinical gene therapy trials and the regulatory issues they raise.

This book provides coverage of the full spectrum of scientific and clinical progress, emphasizing new approaches in the field that currently have the greatest therapeutic application or potential and those areas most in need of future research. Serving both as an introduction to the field of gene therapy and as a general reference, it should prove an invaluable resource for both the expert and new investigator entering the field, as well as the clinician considering enrolling patients in clinical trials.

**Jeffrey Laurence, M.D.**
**August 2014**

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Dr. Laurence received his B.A. Phi Beta Kappa, summa cum laude from Columbia University in 1972, and his M.D. with honors from the University of Chicago Pritzker School of Medicine in 1976. He was elected a Rhodes Scholar to Oxford University in 1973. Deferring this honor, he accepted a Henry Luce Fellowship to Japan, where he worked at the Institute for Cancer Research in Osaka from 1974–1975. Dr. Laurence returned to New York to complete a residency in internal medicine and fellowship in hematology-oncology at The New York Hospital, followed by a research fellowship in immunology at The Rockefeller University.

His work focuses on the mechanisms by which HIV and antiretroviral drugs used in its treatment affect endothelial cells and osteoclasts, in models for thrombosis, cardiovascular disease and osteoporosis linked to HIV. As an outgrowth of this research he is has a long-standing interest in exploring thrombotic microangiopathies associated with complement activating disorders.

Dr. Laurence is a member of several national and international AIDS organizations, and recently received an "Award of Vision" from the Red Ribbon AIDS Foundation. He is also a recipient of the Clinician-Scientist Award of the American Heart Association and the William S. Paley Fellowship in Academic Medicine, and is an elected Fellow of the New York Academy of Sciences and a member of the American Society for Clinical Investigation. He has 3 children and lives in Greenwich, CT.

Michael Franklin, MS is a medical editor in the Division of Hematology, Oncology, and Transplantation (HOT) at the University of Minnesota. He earned his M.S. in science journalism with honors from Boston University in 2000. While in Boston, Mr. Franklin interned at the *Harvard Health Letter* at Harvard Medical School and *Boston Review* at Massachusetts Institute of Technology. He was also a staff writer for *The Daily Free Press*, the Boston University school newspaper, and contributing editor for *Stellwagen Soundings*, a newsletter covering the Stellwagen Bank National Marine Sanctuary. Shortly after graduation, Mr. Franklin began his editorial career as the Managing Editor of *The Journal of Laboratory and Clinical Medicine*, subsequently retitled *Translational Research* in 2006. During his tenure as Managing Editor, Mr. Franklin developed an interest in publication ethics while mediating breaches of scientific misconduct involving authors of the journal. He has written about publication ethics for *Minnesota Medicine* and teaches a seminar on the topic to HOT trainees. Michael has a long-standing interest in the history of science, specifically the history of experimental discoveries in chemistry and medicine, and how scientific reasoning works as an engine of human knowledge. Since becoming a medical editor in 2006, Michael has written or edited hundreds of research reports, grant proposals, book chapters, reviews, educational curricula, and other science-related material for clinicians and scientists. He regularly teaches seminars on writing in the sciences, designing visual displays of data, and how to read a journal article to HOT faculty and/or trainees, as well as other groups, including the *Association of Multicultural Scientists* and the *North Central Chapter* of the *American Medical Writers Association* (AMWA). He served as President of the *North Central Chapter* from 2010–2011 and has been a member of AMWA since 2007. When not writing or editing, Michael spends time with his partner, two daughters, and dog in Minnetonka, Minnesota.

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