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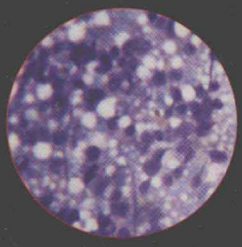
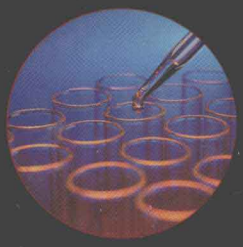

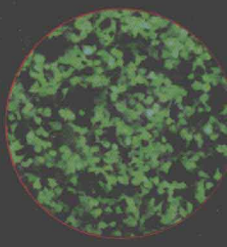

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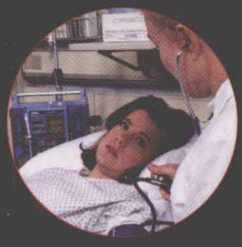

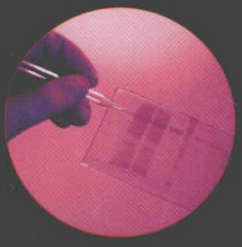
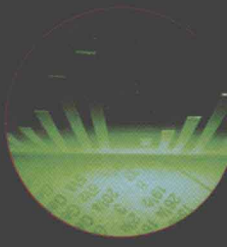




EDITED BY
BORO DROPULIC • BARRIE CARTER

CONCEPTS IN GENETIC MEDICINE

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PREFACE

Our goal in compiling *Concepts in Genetic Medicine* was to produce a book that looked broadly at the issues surrounding the development of gene-based therapies. Consequently, topics include new developments in vector design and methods for their application to various disease states, the manufacture and testing of gene transfer products; understanding the regulatory environment for development of gene therapy products; some considerations as to how the development of gene therapy products financed toward commercialization, and finally how companies commercializing gene therapeutic products will ultimately realize returns on their investment.

The organization of the chapters approximates the chronological order of product development. After an insightful introduction by Theodore Friedmann, we start with vector design by focusing on select examples and their potential utility for the treatment of specific diseases. The selection is by no means exhaustive, but provides the reader with some background as to the major vector classes in development and the specific diseases for which they are being targeted for therapy. The subsequent set of chapters summarize methods for the manufacture and release testing of some of these vectors, emphasizing methods and processes that are relevant for their application in human clinical trials. The following chapters expand on the regulatory theme, providing key concepts for preclinical studies and clinical trial design. The final chapters provide important insights as how to finance the development of gene therapy products using private equity investment as a vehicle, and how the return on investment in these companies will be actualized by new reimbursement strategies for cellular and gene therapy products.

It is important to note that *Concepts in Genetic Medicine* is not designed to be a thorough review of every potential gene therapeutic strategy currently in development in numerous laboratories around the world. Rather, its aim is to provide salient examples of such strategies so that the book can broadly cover many of the *concepts* that need to be taken into consideration when developing gene therapy products: from basic research in the laboratory to full commercialization by companies. We believe that successful development of these new revolutionary products will happen only when these considerations are taken into account early in the product development cycle. Only then will the goals of this field be realized: revolutionary treatments for serious diseases and unmet medical needs where other approaches have failed to provide a satisfactory outcome or cure.

BARRIE CARTER
BORO DROPULIC

CONTENTS

APPLICATIONS

THE MANUFACTURE OF GENE THERAPY PRODUCTS

1 The Evolution of Human Gene Therapy: A Journey from Excessive Hype to Excessive Diffidence to Reality

THEODORE FRIEDMANN
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Unlike Athena, who emerged from the brow of Zeus fully armed and ready for her godly duties, advances in biomedicine are not born fully formed and mature. Virtually all of the therapies and preventive methods that we take for granted—cancer chemotherapy, immunization techniques, tissue transplantation, management of cardiovascular disease, and treatment of diabetes and many other metabolic and degenerative diseases—have required decades of development and incremental advance from initial concept and early proof of concept to truly effective and widely applicable clinical application. They are all still imperfect but are evolving rapidly, and their practitioners are learning from false starts, detours, reversals, and missteps. In most cases, scientists, the public, policymakers, and the media understand and accept what is often a discouragingly slow pace of advance in a difficult new science. In contrast and for many reasons, the field of gene therapy found itself in its early stages on a somewhat unusual path, with many segments of the community—basic and clinical scientists and their institutions, the public and its agents, disease foundations and patients' interest groups, the media, and the biotechnology and pharmaceutical industries—too often expecting immediate success and not appreciating the inevitable need for slow, incremental evolutionary growth.

There have been enough reviews of the history of gene therapy from concept to clinical application to establish the fact that it is still a very young discipline [1,2]. Its most obvious conceptual origins date back no further than the late 1960s and early 1970s [3], and its clinical applications began only in 1989–1990 [4,5]: a clinical history of a mere 15 to 16 years. In that relatively short period of

Concepts in Genetic Medicine, Edited by Boro Dropulic and Barrie Carter

time, the birth and development of the field of human gene therapy have been characterized not only by impressive scientific and medical innovation but also by controversy and missteps that have at times inappropriately overshadowed the impressive scientific and medical achievements that have already begun to convert basic gene transfer technology into truly effective therapy. Fortunately, most of us now understand that human gene therapy is no Athena, but rather, is at the very earliest stages of its evolutionary process. Even so, there have been frequent reminders from parts of the scientific and biomedical communities and from the media that the gene therapy community has largely failed to deliver on its stated and implied promises of rapid and even imminent clinical benefits of gene transfer technology delivered by the gene therapy community itself and by research institutions, disease foundations and funding agencies, and private industry.

Without doubt, the early and often overstated clinical promise of gene therapy has been largely unfulfilled—we can all agree on that. But that fact speaks less to the merits of the scientific and medical results than to the exaggeration and unattainable goals of many early expectations as well as to the extreme difficulty of the task. Disappointment does not occur in a vacuum—it is always a result of unmet expectations. It has obviously been the unrealistic expectations that have been most responsible for the widespread disappointment in the achievements of gene therapy until now. The unrealistic early clinical claims have produced unattainable goals that in turn led to disappointment with the apparent slow pace of clinical success and to the common reflexive preoccupation on the part of many critical observers with the trees and not the forest. What is this forest, and has the field of gene therapy actually achieved something important even so early in its life?

The forest is the fact that a conceptually new form of medicine has been born, that it is still very immature, but that like human infants, it is beginning to show hints of future maturity. A number of recent studies have indicated without any doubt that clinical applications of basic clinical gene transfer studies can indeed improve the course of disease and ameliorate suffering. Such improvements have not been trouble-free and have come at some great cost, but in several instances they have constituted undeniable savings of lives and improvements in quality of life. Without disregarding the reversals and difficulties of the past, that development must be called therapy. Some of the most convincing objective therapeutic results have come in the gene transfer studies of the monogenic immunodeficiency inborn errors of metabolism, a group of several distinct lethal diseases for which therapy has remained largely inadequate. For some of these disorders, such as the inborn errors of metabolism that cause severe combined immunodeficiency disease (SCID), bone marrow transplantation has, when feasible, allowed excellent and even definitive treatment. But for the many patients with one or another form of SCID for whom this option is not available, far less effective symptomatic therapies have been used but generally not with uniform success. The new form of treatment represented by gene transfer studies for several of these disorders, especially X-linked SCID (X-SCID) [6], adenosine deaminase

deficiency SCID (ADA-SCID) [7], and most recently, chronic granulomatous disease (CGD) [8], has allowed patients to achieve virtually full immunological reconstitution and thereby to survive and even thrive for up to and exceeding six years after treatment: to attend school and to roll around in the dirt with their playmates, that is, to lead the perfectly normal childhood lives previously not possible for them.

We are all painfully aware that the treatment has been clouded by the development of a leukemialike disease in three of the children and the death of one child as a direct result of the therapy. So we have relearned the lesson that we have learned from many other early and still developing therapies: that even undeniably effective but imperfectly understood therapeutic procedures can, and do, lead to serious adverse and even lethal consequences. In the case of the X-SCID disease model, we have learned of the technical problems caused by integration of vectors into unforgiving regions of the genome in recipient children, and there is evidence that the X-SCID model itself may be severely complicated conceptually by the possibility that the γ-C gene, the gene responsible for X-SCID and that must be reconstituted in patients, can itself be an oncogene. Fortunately, the results with ADA-SCID have not yet been reported to produce similar adverse consequences, possibly because the ADA gene does not have similar oncogenic properties. There are other tantalizingly promising early results in other human disease settings, including forms of cancer, cardiovascular disease, blindness, and others [9–14].

At the clinical level, this evolution from the first gene transfer studies in human subjects to the present time of unequivocal clinical therapeutic efficacy has taken approximately 16 to 17 years, a remarkably rapid course compared with many technically complex areas of therapy. Therapeutic success stories usually develop slowly and with incremental advances over several decades, usually through stages of severe conceptual and technical setbacks and failures. For instance, the beginnings of antimetabolite treatment for childhood leukemia with the introduction of the folic acid antagonist aminopterin by Sidney Farber and colleagues in 1948 [15] came at a time when successful salvage from childhood T-cell leukemia occurred at a rate below several percent. With additional drug discovery and refinements in delivery, therapeutic success increased inexorably to its current level of 90% or greater, but that change required 30 to 40 years. Similarly, there are numerous other examples of decades-long development and maturation times required for other, now standard forms of therapy to progress from conception to initial glimmerings of treatment success to truly effective and widespread application. Consider, for instance, the histories of cancer chemotherapy, organ and tissue transplantation, and the clinical application of monoclonal antibodies. Every one of these and many other therapies came only after several decades of incremental advances, incorporating lessons learned from many false starts, errors, and setbacks.

Not only does it take time for new concepts to mature into effective therapy, but it also evident that it can be precisely at the time when gene transfer begins to be efficient and therapeutically effective that serious clinical setbacks may

first appear. Consider the well-known induction of secondary tumors during successful and lifesaving chemotherapy and radiotherapy of cancer. It is with increasing efficacy of aggressive treatment that the induction of secondary tumors came to be revealed. These and other types of adverse events may therefore not necessarily represent conceptual errors or flaws in the experimental design so much as the harm inherent in effective, yet imperfect therapy itself. In that regard, the induction of leukemia in some patients in the X-SCID study might be seen to represent harm intrinsic to effective therapy in the same way that secondary tumors are an intrinsic and inevitable consequence of effective but still flawed therapy for cancer. It seems very likely that leukemias or other unwanted consequences for retrovirus-mediated gene transfer studies have not been seen in previous studies at least partially because gene transfer and transgene expression have previously simply been too inefficient. Once gene transfer became efficient enough to permit frequent provirus integration near oncogenes and to lead to stable and efficient expression of the therapeutic transgene, tumorigenesis occurred in transduced cells. In the case of retrovirus vectors, it is difficult at the present time to envision a solution to this problem short of site-specific integration of the transgene, but methods are emerging that begin to make the possibility of definitive sequence correction of mutations through site-specific genetic modification seem feasible [16]. Similarly important new methods are emerging that promise specific control of gene expression through modulation of RNA expression [17].

At the clinical level, gene therapy has had an unusually short history of merely 15 to 16 years, admittedly to enormous publicity and great early academic and commercial expectations of imminent success. However, consider what has occurred in that short time. Not only has this completely theoretical approach to disease treatment established itself as a powerful new concept in medicine, but it has also become a very large worldwide effort in academia and industry. Furthermore it has delivered a handful of results that provide inescapable proof of the concept that human disease can indeed be treated at the level of the underlying genetic defects and not only at the symptomatic or metabolic level. Its course has certainly been irregular and even contentious because of missteps and setbacks, overstated early progress, and therapeutic claims. But the field as a whole has learned well from these experiences and has clearly recognized the need for greater care and rigor than was evident at times during the earliest clinical period of the field of gene therapy. Most investigators understand well the hazards of shortcuts and appreciate that studies in this field of biomedicine should be carried out with all the rigorous care required of other areas of clinical research.

Notwithstanding setbacks and treatment-associated harm, progress has been real, and the time has arrived for a more realistic and sober appreciation of the field of gene therapy. Some critics might well be advised to temper their reflexive preoccupation with past difficulties with a more realistic recognition of the important advances in the field and of the undeniable clinical benefits in some studies. Just as important, it is an appropriate time for proponents and advocates of gene therapy to put aside what has become almost timidity in the face of the admitted difficulties and setbacks and begin to point more effectively

to the real successes and achievements of the field—to point justifiably to the important achievements in the field and to do so with an appreciation not only of the conceptual and technical missteps of the past but also of the great conceptual and technical advances that have been made.

As this volume attests, gene therapy is no will-of-the wisp and no mirage, either as a stand-alone approach to treatment of some disorders or as adjunct treatment for many other common and widespread disorders, such as most forms of cancer. Those who have conceived and shaped this field and who are working to bring it to the relief of illness have good reason to be pleased with the recent progress and with the future promise.

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