

METHODS IN MOLECULAR BIOLOGY

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Electroporation Protocols

Preclinical and Clinical Gene Medicine

Second Edition

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Preface

Electroporation gene therapy, or gene electrotransfer, refers to delivery of genetic materials into target tissues or cells via electric pulses for treatment or prevention of disease. The genetic materials delivered by electroporation include oligoDNA, RNA, genes, and chromosomes. This term is a by-product of electrochemotherapy, in which chemical therapeutics or synthetic small molecules are delivered into targeted cells via electric pulses. Simultaneous delivery of both genetic materials and chemical therapeutics is referred to as electrochemogene therapy. Delivery of electric signal alone, without any chemical therapeutic or genetic material, is used to deplete tissue (such as tumor tissue), a technique referred to as irreversible electric therapy. The focus of this book is to provide in-depth knowledge and hands-on protocols for delivery of naked DNA and small-interfering RNA (siRNA) to the targeted cells, tissues, and animals for prevention and treatment of disease. Therefore, this book is primarily dedicated to electroporation gene therapy.

The first gene delivery via electroporation was carried out in 1982 by Professor Eberhard Neumann using an *in vitro* cell culture system. During the more than 30 years since then, this technology has evolved greatly thanks to the remarkable progress in genetic sequencing, gene array analysis, gene cloning, gene expression detection, DNA manufacture, and discovery and synthesis of siRNA. This second edition of *Electroporation protocols: Preclinical and Clinical Gene Medicine* builds on the success of the first edition and on the progress made in siRNA delivery and DNA vaccines for large animals as well as discovery of electroporation applications for the fragile tissues and for internal organs.

To help investigators who are new to this exciting technology, Part I of this book includes reviews covering the theory of how DNA is delivered into cells and tissues via electroporation. For those who are familiar with this field but anxious to seek ways to boost gene delivery via electroporation, to apply this technology to internal organs and fragile tissues, and to translate this approach to large animals and clinical studies, Parts I, II, V, and VI of the book provide reviews and protocols addressing each of these topics.

Because of the rapid development in electroporation gene therapy, 90 % of the chapters in this second edition are completely new; only a few chapters are retained from the previous edition, and they have been heavily revised. Therefore, this book may be considered a continuation of the first edition rather than a replacement in terms of the protocols and reviews included. However, the style and format used for the second edition are similar to those of the first edition and will be very familiar and accessible to investigators who have used the first edition.

The major differences between this edition and the first edition are the additions of a large section on siRNA delivery (Part II) and a section on gene delivery to fragile tissue and internal organs (Part III). The other major differences are the substantial expansion of the section on applications in large animals, primates, and humans (Parts IV and V); the substantial expansion of the section on the use of nonelectric factors to boost gene transfer via electroporation (Part I); the addition of chapters on stem cell gene transfer via electroporation; and the substantial expansion of sections on DNA vaccines (Part V), which may be the

first FDA-approvable clinical product of electroporation gene therapy. The expansion of the large animal and clinical application chapters will help investigators borrow from these successes in promoting their own clinical applications within their own areas of expertise. The new siRNA chapters should assist investigators in using this simple but competitive technology to study gene function in vitro and in vivo without concern for the nonspecific effects of siRNA transfection, such as inflammation signal activation or gene expression when other transfections agents are used. These additions and expansions in the second edition, unfortunately, required sacrifice of chapters of the first edition related to cell-based gene transfer and to electric device description. For details of those topics, the first edition should serve very well.

In summary, this second edition aims to provide comprehensive coverage of the basic theory and practical application of electroporation siRNA therapy, gene therapy, and vaccine. It aims to provide the most current views from the experts in this field; to speed up the understanding and application of electroporation gene transfer; and to provide a valuable and up-to-date resource for investigators both inside and outside this field. A chapter on future applications, specifically irreversible electroporation, is also included.

This editor would like to acknowledge the coeditors of this book, Drs. Richard Heller, Justin Teissie, and Jeffrey Cutrera, who assisted in organizing the book, soliciting chapter authors, contributing their views, and finalizing the contents. I also am grateful to all the authors who worked so hard to get this book to press in a timely manner. Finally, I am very grateful to my department chair, Eugenie Kleinerman, M.D., for providing the space and time I needed to accomplish this work.

Houston, TX, USA

Shulin Li

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