

SPRINGER BRIEFS IN BIOCHEMISTRY AND  
MOLECULAR BIOLOGY

Gerhard Bauer  
Joseph S. Anderson

# Gene Therapy for HIV

## From Inception to a Possible Cure



Springer

# SpringerBriefs in Biochemistry and Molecular Biology

For further volumes:

<http://www.springer.com/series/10196>



Gerhard Bauer • Joseph S. Anderson

# Gene Therapy for HIV

From Inception to a Possible Cure

 Springer

Gerhard Bauer  
University of California Davis  
Sacramento, CA, USA

Joseph S. Anderson  
University of California Davis  
Sacramento, CA, USA

ISSN 2211-9353

ISBN 978-1-4939-0433-4

DOI 10.1007/978-1-4939-0434-1

Springer New York Heidelberg Dordrecht London

ISSN 2211-9361 (electronic)

ISBN 978-1-4939-0434-1 (eBook)

Library of Congress Control Number: 2014930434

© Gerhard Bauer and Joseph S. Anderson 2014

This work is subject to copyright. All rights are reserved by the Publisher, whether the whole or part of the material is concerned, specifically the rights of translation, reprinting, reuse of illustrations, recitation, broadcasting, reproduction on microfilms or in any other physical way, and transmission or information storage and retrieval, electronic adaptation, computer software, or by similar or dissimilar methodology now known or hereafter developed. Exempted from this legal reservation are brief excerpts in connection with reviews or scholarly analysis or material supplied specifically for the purpose of being entered and executed on a computer system, for exclusive use by the purchaser of the work. Duplication of this publication or parts thereof is permitted only under the provisions of the Copyright Law of the Publisher's location, in its current version, and permission for use must always be obtained from Springer. Permissions for use may be obtained through RightsLink at the Copyright Clearance Center. Violations are liable to prosecution under the respective Copyright Law.

The use of general descriptive names, registered names, trademarks, service marks, etc. in this publication does not imply, even in the absence of a specific statement, that such names are exempt from the relevant protective laws and regulations and therefore free for general use.

While the advice and information in this book are believed to be true and accurate at the date of publication, neither the authors nor the editors nor the publisher can accept any legal responsibility for any errors or omissions that may be made. The publisher makes no warranty, express or implied, with respect to the material contained herein.

Printed on acid-free paper

Springer is part of Springer Science+Business Media ([www.springer.com](http://www.springer.com))

# Preface

Combination antiretroviral therapy (ART) for HIV has, without a doubt, saved many lives of people infected with HIV over the last decades, but we are still left with the devastating memory of the large number of deaths caused by HIV and AIDS during the 1980s and early 1990s. To this day, there is still no cure for HIV, and there is also no vaccine to reliably prevent HIV infection. The World Health Organization estimated the number of people infected with HIV worldwide in 2010 at 34 million with about 2.7 million newly infected individuals. This may be an underestimate, however, as many cases go unreported. The total number of AIDS deaths in 2010 was estimated to be around 1.8 million. One of the drawbacks of ART is the fact that it needs to be taken under high compliance lifelong; otherwise the virus will rebound, and drug-resistant mutants can arise. Additionally, many patients suffer from side effects caused by antiretroviral medication, ranging from mild to severe, sometimes limiting quality of life. Another aspect is the high cost of lifelong drug treatment and the limited ability to make these drugs available in developing countries, very much affected by HIV and AIDS. Taken together, these facts have been motivating us and many of our colleagues to continue working towards a cure for HIV that we hope we can elicit with stem cell gene therapy for HIV.

In 1988 an article published by David Baltimore in the journal *Nature* introduced the term “intracellular immunization,” which meant engineering HIV target cells to become resistant to HIV by the insertion of “anti-HIV genes.” Since these early times, when gene therapy was a very new and emerging research field, members of our research group have been working on the development of such a gene therapy application, particularly stem cell gene therapy for HIV. In spite of drawbacks and funding problems, this research has continued to this day, with current developments promising to come much closer to a cure for HIV than ever before.

Several years ago, the idea of a functional cure for HIV was postulated. This was precipitated by an interesting, anecdotal clinical case in Berlin, Germany. An HIV-positive patient with leukemia had to receive an allogeneic bone marrow

transplantation to successfully cure his leukemia. The bone marrow donor had been specifically selected for this patient, not only for the tissue match but also for being homozygous for the CCR5 deletion, a natural chemokine receptor deletion on blood cells, including HIV target cells, that leads to HIV resistance. The phenomenon that a small number of people living in central and northern Europe are carrying this deletion without an adverse phenotype and exhibit natural resistance to HIV is well known. CCR5 acts as the secondary receptor for macrophage tropic strains of HIV, and the absence of this receptor restricts HIV from attaching to and entering HIV target cells. Additionally, most initial HIV infections occur through macrophage tropic strains. After the allogeneic bone marrow transplantation, the patient's leukemia was cured, as expected, but also another remarkable phenomenon occurred: For the last 7 years, there has been no detectable HIV viral load in this patient, under complete ART withdrawal. This case suggests that the transplantation of HIV-resistant hematopoietic stem cells was able to generate an HIV-resistant immune system that has been able to control HIV replication for several years. This transplant is very similar to what has been attempted in stem cell gene therapy clinical applications, and by applying the optimal settings, we believe that the outcome seen in the "Berlin patient" can be repeated using engineered autologous hematopoietic stem cells in other HIV-infected individuals.

In this book, we describe the individual aspects of gene therapy for HIV, from its early development to our current knowledge, from early clinical trials to current and planned future clinical applications, set out to possibly cure HIV. It is the authors' belief that stem cell gene therapy for HIV, if proven successful, can be commercialized and made affordable. Automated, closed system culture systems could be developed, and there is no real technical limit to bringing such systems to developing nations in an easy-to-use application, as long as there is enough effort made to actually develop this. A true impact could also be made if gene therapy vectors could be developed that target hematopoietic stem cells *in vivo*, making them resistant to HIV. Additionally, with this book, it is our sincere wish to also inspire young researchers to take on the interesting field of gene therapy for HIV and help bring about a long-needed cure for the disease.

This book would not have been possible without decades of hard work from many noted researchers in this field. We therefore would like to particularly acknowledge the contributions of Dr. Donald Kohn, Dr. John Zaia, and Dr. John Rossi. The first potent anti-HIV genes applied clinically and the first stem cell gene therapy clinical trials for HIV were initiated by them at City of Hope and Children's Hospital Los Angeles, among them was the first case of a pediatric clinical trial of stem cell gene therapy for HIV. We also would like to thank our colleague Steve Tobin for his valuable input on this manuscript.

# Contents

<b>1 Principles of Gene Therapy</b>	1
Injection of Naked DNA	2
Nonviral Gene Transfer (Transfection) Methods	3
Viral Gene Transfer (Transduction) Methods	4
Retroviruses	5
Other Viruses	7
<b>2 History of Gene Therapy</b>	9
<b>3 Principles of HIV Gene Therapy</b>	17
<b>4 Gene Therapy Vectors</b>	27
Lentiviral Vectors	28
Other Modifications and Additions	30
Anti-HIV Genes	30
RNA Strategies	32
Vector Production	32
<b>5 Stem Cells for HIV Gene Therapy</b>	35
Pluripotent Stem Cells	38
<b>6 Animal Models Used in HIV Gene Therapy</b>	41
Murine Models	41
HIV Infections in Humanized Mice	44
Nonhuman Primates	46
<b>7 Manufacturing of a GMP Grade Product for HIV Gene Therapy</b>	49