Historical Perspective on the Current Renaissance for Hematopoietic Stem Cell Gene Therapy

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KEYWORDS

• Gene therapy • Hematopoietic stem cells • Gammaretroviral vector
• Lentiviral vector • Gene editing • Site-specific endonucleases
• Homologous recombination

KEY POINTS

- Gene therapy using gammaretroviral vectors into hematopoietic stem cells led to immune reconstitution for several primary immunodeficiency disorders, but produced leukoproliferative complications in some cases.
- Lentiviral vectors have become the predominant gene addition tool for hematopoietic stem cells and are showing efficacy and safety for numerous genetic blood cell diseases.
- Gene editing through the use of site-specific endonucleases is an emerging technology that may be applied for gene therapy using hematopoietic stem cells.

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Disclaimer: Due to word limits, only essential references are included.

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IN THE BEGINNING

It took more than 3 decades, but now gene therapy for blood cell diseases has become a clinical reality, as documented in the articles of this issue. From a bold vision proclaimed in editorials in the 1970s to 1980s,[1,2] steady incremental improvements in the underlying technology and successive series of clinical trials have advanced the state of the field so that a first hematopoietic stem cell (HSC)-based gene therapy product has been licensed for marketing in the European Union, with several more likely to follow there and in the United States.

The origins of gene therapy lie within the origins of modern molecular biology. The capacity to clone human genes as complementary DNA (cDNA) led to isolation of clinically relevant genes such as *beta-globin*, *HPRT*, *ADA*, and *DHFR*. Gene transfer to mammalian cells began with the development of calcium phosphate transfection methods, using selectable marker genes such as HSV *TK* and *DHFR*.[3] Early viral vectors derived from SV40 and Polyoma were also examined. Plasmids carrying the HSV *TK* gene or a human *beta-globin* cDNA were transfected into murine bone marrow (BM) cells and detected in vivo after cells were reinjected into mice.[4]

The field of gene therapy for blood cell disorders suffered an early setback when an initial attempt was made to translate the results from murine studies to clinical gene therapy for beta-thalassemia. The method used was ex vivo transfection of BM cells using a plasmid containing a human beta-globin cDNA. Two patients with beta-thalassemia major with severely advanced clinical complications were treated.[5] No clinically beneficial effects were detected, but no adverse effects resulted either (these results were never published). The study was performed in Italy and Israel at a time when there was no institutional review board approval to perform the studies at the primary US institution, due to concerns about insufficient preclinical efficacy data. The subsequent investigations led to censure of the lead scientist.[6]

The history of gene therapy cannot be fully told without a tribute to the role that the National Institutes of Health (NIH) Recombinant DNA Advisory Committee (RAC) played for the clinical translation of gene therapy, at least in the United States.[7] The RAC was formulated at NIH in the mid-1970s to provide guidance on policies to protect public safety as recombinant DNA technology was emerging, especially as potential biohazards were addressed by the scientific community at the landmark Asilomar meetings.[8] Following the violation of rules in the initial beta-globin gene transfer study, the Human Gene Therapy Subcommittee (HGTS) of the RAC was tasked by NIH Director Donald Fredrickson with formulating rules for review and conduct of federally funded clinical trials of gene therapy. Over the next decade, the HGTS developed a process for applying for permission from the NIH to perform clinical investigations of gene therapy. The existence of this highly rational and well-considered body obviated passage of legislation to govern this activity. The RAC played a very active role in shaping the conduct of gene therapy trials in the early days under public purview. Its role has been successively rolled back over time as greater experience was gained with clinical gene therapy, and the RAC has increasingly focused on novel issues of biosafety, and less on details of clinical trial design and performance.

GAMMARETROVIRAL VECTORS EMERGE

In the 1980s, recombinant versions of murine gammaretroviral vectors (gRV), primarily derived from the Moloney murine leukemia virus, were developed to carry genes into mammalian cells.[9,10] Several studies followed demonstrating that these vectors were capable of introducing foreign genetic material, often in the form of the neomycin phosphotransferase marker gene into HSC in murine BM cells.[11–13] Transplantation of gRV-treated murine BM led to the production of mature blood cells containing