Gene Therapy (2003) 10, 933–934

www.nature.com/gt

INTRODUCTION TO THE SPECIAL ISSUE

Virology and immunology of gene therapy, or virology and immunology of high MOI infection with defective viruses

PR Lowenstein[1,2]

[1]Gene Therapeutics Research Institute, Cedars-Sinai Medical Center, Research Pavilion, 8700 Beverly Boulevard, Suite 5090, Los Angeles, CA 90048, USA, and [2]Department of Medicine, David Geffen School of Medicine, University of California Los Angeles, Los Angeles, CA, USA

Gene Therapy (2003) **10,** 933–934. doi:10.1038/sj.gt.3302073

"All authors in this volume are realists – you will not find them talking about fictitious things. But one has to exercise caution because as with all discussions about reality, it always pays to be aware of my sixth law of thermodynamics, never falsified: things aren't as they seem"

Joel Cracraft

In "Species concepts in theoretical and applied biology: a systematic debate with consequences"; In: Species Concepts and phylogenetic theory. A Debate (Eds. Quentin D. Wheeler and R. Meier) Columbia University Press, NY, 2000, p. 3–14

The conversion of pathogenic viruses into vectors for therapeutic genes is avowedly a simple idea. Any virus is a potential vector for gene delivery if two simple maneuvers can be completed: the elimination of virus replication (or, its capacity to cause disease), and the insertion of a gene of interest (or, its capacity to deliver a therapeutic gene).[1]

These two maneuvers have now been accomplished for many viruses, both in experimental paradigms and in clinical trials. However, one impediment remains to be overcome: the immune system. The innate and adaptive arms of the immune systems have evolved over millions of years to detect miniscule quantities of foreign agents within the body. Work during the last 5 years has even highlighted that initial viral interactions with membrane receptors signals their presence to the about-to-be-infected cells.[2–4] As a consequence, virions impaired for entry into cells and subsequent target cell transduction can still stimulate proinflammatory intracellular signaling cascades and the priming of adaptive immune responses.

The challenges imposed by all arms of immune surveillance have stimulated further improvements in viral vectors, beyond the elimination of virus replication and encoding of transgenes. Advanced vector systems available are derived from viral genomes that have been deleted of all sequences encoding viral proteins. The only viral nucleic acid remaining represents sequences required for genome replication and packaging into virions.[5,6] Further, capsids have been engineered to reduce viron binding to endogenous viral receptors, and to retarget infection of predetermined cell types.[7] Transcriptional regulatory systems facilitate directed expression of transgenes in a cell-type-specific fashion.[8] In some instances, immunogenicity of the transgene product itself can be reduced.

However, compared to the low amount of infecting virions required to establish a normal viral infection, nonreplicating viral vector virions must be delivered in substantially higher quantity to achieve significant levels of therapeutic gene expression. Thus, even though these vectors encode no viral proteins, large numbers of viral capsid proteins enter the cell. In addition, even if the immune system does not respond normally to an endogenously expressed protein, its *de novo* expression from a viral vector in a novel cell type could lead to an immune response perhaps due to different processing of the therapeutic protein that may make available novel antigenic epitopes, which, in combination with vector-induced inflammation, could lead to an immune response.

An understanding of the quantity and quality of immune responses against viral vectors will pave the way toward novel approaches to overcome this last barrier so that efficient, effective, and safe gene transfer can be achieved in the clinic.

This special issue of gene therapy, immune responses to viral vectors, explores the problems posed by the immune system. Especially, this issue surveys the inflammatory and immune responses to HSV-1 vectors,[5] AAV-vectors,[9,10] adenovirus vectors,[6,9,11] and further investigates how different arms of the immune response affect both replication-defective[5,6,9] and replication-competent vectors.[5,12,13] Of further importance to the field, this issue explores how inflammatory and immune responses will vary, even for the same vector, depending on the target organ being injected and transduced.[6,11,13]

Thus, this special issue examines immune responses to vectors infecting different tissues, that is, muscle,[9] liver,[10] eye,[11] and brain.[6] Main themes of this issue include early inflammatory responses and signaling pathways activated by viral vectors;[4] how viral vectors stimulate the release of inflammatory mediators, cytokines, and

chemokines;[13] how the inflammatory response elicited depends on vector components (eg viral capsids, transgenes, marker genes), viral vector dose, site of injection, tissue injected, and cells transduced;[4,6,9] how different vectors encoding a similar transgene, but injected into different tissues, can stimulate very different types of immune responses;[9] the mechanisms by which the innate and/or adaptive arm of the immune response can inhibit transgene expression (or eliminate transduced cells).[6,12,13] Clinical immune responses have not been discussed, since they were reviewed (at least those concerning adenoviral vectors) in detail elsewhere.[14]

While the astute reader will find no quick answers on how to 'get away with it', that is, forgetting the immune response continues to impose a major theoretical and practical hurdle to the safe implementation of clinical gene therapy, the goal is to share with the larger gene therapy community, the possibility that by understanding the molecular and cellular mechanisms underlying the activation of immune responses by viral vectors, and the elucidation of the pathways by which activated arms of the inflammatory and immune pathways interact with transduced cells, the field will be on the high ground to design better and safer vectors, and implement more effective and compelling clinical trials.

Acknowledgements

I wish to thank Lynn W Enquist for suggestions and continuous illuminating discussions on the interface between gene therapy and virology.

References

1 Kay MA, Glorioso JC, Naldini L. Viral vectors for gene therapy: the art of turning infectious agents into vehicles of therapeutics. *Nat Med* 2001; **7**: 33–40.
2 Kurt-Jones EA *et al*. Pattern recognition recptors TLR4 and CD14 mediate responses to respiratory syncytial virus. *Nat Immunol* 2000; **1**: 398–401.
3 Bieback K *et al*. Hemmagglutinin protein of wild-type measles virus activates toll-like receptor 2 signaling. *J Virol* 2002; **76**: 8729–8736.
4 Liu Q, Muruve DA. The molecular basis of the inflammatory responses to adenovirus vectors. *Gene Therapy* 2003; **10**: 935–940.
5 Bowers WJ, Olschowka JA, Federoff HJ. Immune responses to replication-defective HSV-1 type vectors within the CNS: implications for gene therapy. *Gene Therapy* 2003; **10**: 941–945.
6 Lowenstein P, Castro MG. Inflammation and adaptive immune responses to adenoviral vectors injected into the brain: peculiarities, mechanisms, and consequences. *Gene Therapy* 2003; **10**: 946–954.
7 Wickham TJ. Ligand-directed targeting of genes to the site of disease. *Nat Med* 2003; **9**: 135–139.
8 Castro M *et al*. Regulatable and cell-type specific transgene expression in glial cells: prospects for gene therapy for neurological disorders. *Prog Brain Res* 2001; **132**: 655–681.
9 Joos K, Chirmule N. Immunity to adenovirus and adeno-associated viral vectors: Implications for gene therapy. *Gene Therapy* 2003; **10**: 955–963.
10 Sun JY, Anand-Jawa V, Chatterjee S, Wong Jr KK. Immune responses to adeno-associated virus (AAV) and its recombinant vectors. *Gene Therapy* 2003; **10**: 964–976.
11 Bennett J. Immune responses following intraocular delivery of recombinant viral vectors. *Gene Therapy* 2003; **10**: 977–982.
12 Wakimoto H, Johnson PR, Knipe DM, Chiocca EA. Effects of innate immunity on herpes simplex-virus and its ability to kill tumor cells. *Gene Therapy* 2003; **10**: 983–990.
13 Chen D, Murphy B, Sung R, Bromberg JS. Adaptive and innate immune responses to gene transfer vectors: role of cytokines and chemokines in vector function. *Gene Therapy* 2003; **10**: 991–998.
14 See reviews in: *Hum Gene Ther* 2002; **13**: 1–175.