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Jan VAN. SLAMBROUCK, MD, Marianne S. CARLON, PhD, Dirk VAN. RAEMDONCK, MD, PhD, Laurens J. CEULEMANS, MD, PhD

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Commentary: Genetic modulation in lung transplantation: Epic odyssey of vector transduction and transgene expression

Jan VAN SLAMBROUCK, MD\textsuperscript{1,2}; Marianne S. CARLON, PhD\textsuperscript{2,3}; Dirk VAN RAEMDONCK, MD, PhD\textsuperscript{1,2}; *Laurens J. CEULEMANS, MD, PhD\textsuperscript{1,2}

\textsuperscript{1}Department of Thoracic Surgery, University Hospitals Leuven
\textsuperscript{2}Department of Chronic Diseases and Metabolism, Laboratory of Respiratory Diseases and Thoracic Surgery (BREATHE), KU Leuven University, Leuven, Belgium
\textsuperscript{3}Department of Pharmaceutical and Pharmacological Sciences, Molecular Virology and Gene Therapy, KU Leuven University, Leuven, Belgium

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*Corresponding Author Contact Information:

Laurens J. Ceulemans, MD, PhD
Department of Thoracic Surgery and Lung Transplantation
University Hospitals Leuven
Herestraat 49, B-3000 Leuven, Belgium
E-mail: laurens.ceulemans@uzleuven.be; Phone: +32 16 34 6820  Fax: +32 16 34 68 21
Central Message: The field of lung transplantation embarks on an epic odyssey to explore the feasibility of using the adeno-associated viral vector for genetic modulation to improve the short- and long-term outcome.

Central Picture Legend: Lung transplant research embarks on an odyssey in search of genetic donor lung modulation. Picture publicly accessible at:


Figure 1 Legend: Lung transplant researchers are at the start of an odyssey of vector transduction and transgene expression aiming towards clinical use. Many challenges lie ahead before we can reach the safe harbor of Ithaka. Poem "The City" from C.P. Cavafy: Collected Poems. Translated by Edmund Keeley and Philip Sherrard. Publicly accessible at:

https://www.poetryfoundation.org/poems/51296/ithaka-56d22eef917ec

1 Genetic lung modulation is a novel strategy to overcome the challenges that hamper further improvement of lung transplantation (LTx) outcome.

2 We congratulate the authors for embarking on this odyssey in search of an efficient delivery vector and route. Previously, in the field of LTx, adeno- and lentiviral vectors have been investigated.\(^1\)\(^2\) However, the adeno-associated viral (AAV) vector is characterized by lower pathogenicity, lower risk for genomic integration, stable transgene expression, and existing clinical applications.\(^3\)

3 Kesseli and colleagues (Duke University, NC) assessed for the first time in a syngeneic rat model of orthotopic left-LTx the potential to genetically target the donor lung ex-vivo with AAV.\(^4\) A low/high dose (\(8e^{10}/4e^{11}\) copies) of AAV9 was delivered ex-vivo in the bronchus or
pulmonary artery, prior to 60 minutes of static cold storage (SCS). Reporter gene expression was assessed with immunohistochemistry after two weeks (Green Fluorescent Protein, mCherry) and in-vivo or tissue luminescence (Luciferase) up to two months after delivery. Bronchial delivery resulted in epithelial expression. Arterial instillation did not result in clear endothelial expression but lead to off-target expression in liver and heart.\(^4\)

Viral vector transduction in LTx remains uncharted territory. Every step of this odyssey undertaken by the transgene and its vector, from delivery up to the target cell, requires our detailed understanding. Many challenges lie ahead:

Regarding the vector, targeting a specific cell requires a vector with a well-considered cell tropism and delivery route. A wide range of AAV serotypes and capsid variants targeting various cell types is available.\(^3,5\) Due to the rate-limiting second strand synthesis, transgene expression after AAV delivery with a single-stranded genome (ssAAV) typically begins after 1-2 days reaching maximal expression at 2-4 weeks.\(^6,7\) Synthetic vectors like lipid nanoparticles are a non-viral alternative. Their advantage is that they are even less immunogenic and can be loaded with mRNA, enabling more rapid expression kinetics.\(^8,9\)

Through bronchial delivery, the vector is directly exposed to the large surface area of bronchiolar and alveolar epithelium. Arterial instillation theoretically favors endothelial transduction but may potentially result in more systemic spread and subsequent off-target effects. In addition, reliably determining the cells that are transduced is pivotal to determine vector tropism. We encourage the use of multiplex fluorescent immunohistochemical labelling to co-localize reporter genes and cell type markers.

Delivery during SCS is technically less demanding but ex-vivo lung perfusion (EVLP) might provide a beneficial effect on the efficiency of vector cell-entry and transgene expression. EVLP allows ex-vivo delivery of the vector while cell metabolism is maintained.
Delivery through the perfusate might provide a prolonged and more intricate contact with the target cells. Rodent models will allow further investigation of vector delivery during EVLP.10

Gene therapy is already pushing the therapeutic boundaries for many diseases.3 Also the LTx research community should set sail towards clinical implementation of genetic modulation for improvement of LTx outcomes. Preparing for the challenges ahead, by learning from gene therapy applied in other medical fields, will be key to reach the safe harbor of Ithaka, as depicted in the poem by C.P. Cavafy (Figure 1).


Keep Ithaka always in your mind. Arriving there is what you’re destined for. But don’t hurry the journey at all. Better if it lasts for years, so you’re old by the time you reach the island, wealthy with all you’ve gained on the way, not expecting Ithaka to make you rich.

Ithaka gave you the marvelous journey. Without her you wouldn't have set out. She has nothing left to give you now.

And if you find her poor, Ithaka won’t have fooled you. Wise as you will have become, so full of experience, you’ll have understood by then what these Ithakas mean.

"The City" from C.P. Cavafy: Collected Poems. Translated by Edmund Keeley and Philip Sherrard.