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

Jan Van Slambrouck, MD, a,b Marianne S. Carlon, PhD, b,c Dirk Van Raemdonck, MD, PhD, a,b and Laurens J. Ceulemans, MD, PhD, a,b

Genetic lung modulation is a novel strategy to overcome the challenges that hamper further improvement of lung transplantation (LTx) outcome. 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 Kesseli and colleagues4 from Duke University 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. A low/high dose (8e10/4e11copies) of AAV9 was delivered ex vivo in the bronchus or pulmonary artery, before 60 minutes of static cold storage. Reporter gene expression was assessed with immunohistochemistry after 2 weeks (green fluorescent protein, mCherry) and in vivo or tissue luminescence (Luciferase) up to 2 months after delivery. Bronchial delivery resulted in epithelial expression. Arterial instillation did not result in clear endothelial expression but led to off-target expression in liver and heart.5 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 typically begins after 1 to 2 days, reaching maximal expression at 2 to 4 weeks.6,7 Synthetic vectors like lipid nanoparticles are a nonviral 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 labeling to colocalize reporter genes and cell type markers.
Delivery during static cold storage 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 toward 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).

References