In Vivo Electroporation-Mediated, Intrahepatic Alpha1 Antitrypsin Gene Transfer Reduces Pulmonary Emphysema in Pallid Mice

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Introduction

Alpha1 Antitrypsin (AAT) deficiency is an autosomal codominant disease leading to lung and liver damage. Current therapeutic options are comprised of lung or liver transplantation, or life-long intravenous weekly augmentation therapy. We tested electroporation-mediated AAT gene therapy in the liver in mice that carry a mutation in the palloid gene.

Materials and methods

Electroporation-mediated gene transfer was performed on C57BL/6J-backcrossed palloid mice (pa/pa) using Plasmids pEF-AAT carrying the complete human SERPINA. After performing lung function tests on a flexiVent system (SCIREQ) the mice were sacrificed in order to perform further analysis on bronchiolalveolar lavage, blood, lung and liver tissue.

Results

Figure 1 Immunofluorescent staining depicted the expression of humanAAT 30 days after intrahepatic electroporation-mediated AAT gene transfer by mice liver cells with the anticipated cytoplasmatic distribution of the protein.

Figure 2 Lung function tests showed that 30 days after AAT gene transfer to palloid mice, static compliance was reduced to the level of wildtype mice (Fig. 2a). Pallid mice displayed a significant reduction in lung capacity after AAT gene transfer, depicting values in the range of healthy wildtype mice (Fig. 2b). After AAT gene transfer, hysteresis was reduced towards the range observed in healthy wildtype mice (Fig. 2c). Consistent with these results, palloid mice exhibited an upward and leftward shift in PV relation compared to wildtype mice and compared to mice receiving AAT gene transfer; significant differences were found in the area between the inflation and deflation limb of the PV curves (Fig. 2d).

Figure 3. Left lobe lung histology after intrahepatic AAT gene transfer by electroporation in vivo. Representative micrographs of the lungs of animals from different groups. (a) Normal lung from wildtype (WT) (C57BL/6J) mice, (b) air space enlargement visible in the non-transfected palloid mice and (c) in the animals treated with empty vector, (d) animals treated with the pEF-AAT plasmid: × 20 magnification, scale bar 500 μm. Inset, images at higher magnification as indicated by dashed frame (digital zoom; scale bar 200 μm).

Figure 4. Lung morphometry: (a) septal surface area and (b) mean linear intercept (LM) in different experimental groups, as indicated. For each animal, the left lobe was analysed, and all parameters relate to this specific lobe. Statistical values are expressed as mean ± SD, unpaired t test was performed, and each group was compared to each other. * p < 0.05, ** p < 0.01.

Conclusion: Intrahepatic electroporation-mediated AAT gene transfer is a feasible, safe, and reproducible approach in the context of a mouse model of AAT deficiency with spontaneous pulmonary emphysema. In addition to improved lung function, electroporation-mediated AAT gene transfer effectively reduces local neutrophil activity. While the current study offers a good model for correction of lung damage in AAT deficiency, a more elaborate preclinical study is required using large animal models before clinical translation is considered.

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