Lentivirus GM-CSF gene therapy for autoimmune pulmonary alveolar proteinosis

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Autoimmune pulmonary alveolar proteinosis (aPAP), is a lung disease characterised by the accumulation of surfactant, leading to respiratory failure, due to anti-granulocyte-macrophage colony-stimulating factor (GM-CSF) auto-antibodies. The standard of care is repeated whole lung lavage (WLL) resulting in temporary remission of symptoms. However, this procedure is invasive, carries risks and can only be performed in highly-specialised centres. Administration of recombinant GM-CSF outcompetes the auto-GM-CSF antibodies and stimulates clearance of pulmonary surfactant by alveolar macrophages, but this treatment is expensive and not available to all patients. Therefore, we hypothesised that rSIV.F/HN, a lentiviral vector specifically pseudotyped for efficient gene transfer to the respiratory epithelium, carrying (m)GM-CSF cDNA, may be of therapeutic benefit. We produced high-titer rSIV.F/HN carrying murine (m)GM-CSF (≥1e9 TU/ml) using scalable, serum-free suspension cultures. Vector transduction of A549 cells (MOI 0.1–1000) led to dose-related expression of mGM-CSF which, compared to recombinant mGM-CSF protein, showed similar activity in a mouse myeloid FDC-P1 cell proliferation assay. Transduction of air-liquid interface cultures (9e6 TU/ALI) resulted in stable, long-term (>6months) expression of mGM-CSF. Transduction of mice (1e7 TU/mouse, $n = 3$/group) generated significant ($p < 0.05$) high levels of mGM-CSF in both lung (treated: median 825 (range 460–3790) pg/ml; control: 0.1pg/ml) and bronchoalveolar lavage fluid (BALF) (treated: 3330 (range 2307–7958) pg/ml; control: 0.1pg/ml) samples. Characterisation of GM-CSF-knockout mice showed that the model recapitulates human disease phenotype, with significantly ($p < 0.05$) increased BALF turbidity and phosphatidylcholine and surfactant protein D levels in BALF and lung. This data provides the foundation for our next study; lentiviral-mGM-CSF-transduction of our knockout mice.