COMPARING THE TRANSDUCTION EFFICIENCY OF A LIQUID BOLUS AND AEROSOL DELIVERED LENTIVIRAL VECTOR FOR CYSTIC FIBROSIS LUNG GENE THERAPY

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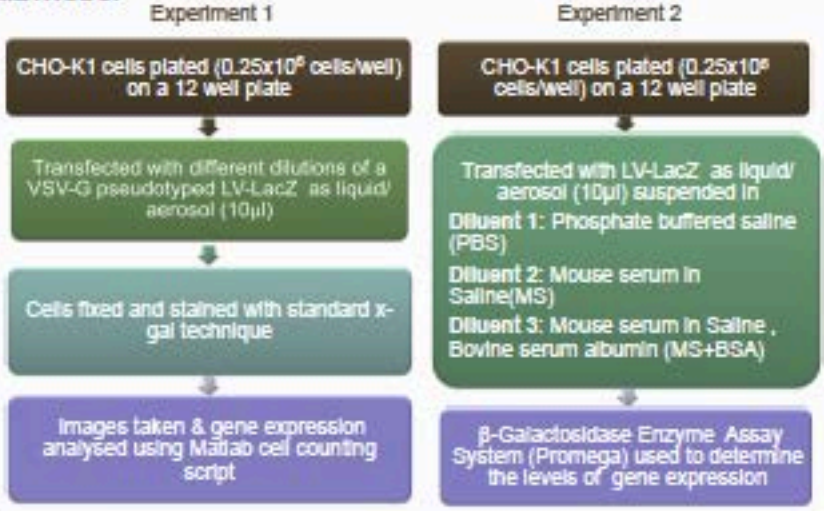
BACKGROUND:

Gene therapy is a potential treatment for cystic fibrosis (CF) lung disease, whereby the therapeutic gene is delivered to the lung to produce functional correction. Aerosol delivery of a gene vector to the lung is an ideal treatment approach because it is non-invasive, easy to administer and less cumbersome compared to liquid delivery. In this study we have carried out *in-vitro* experiments to test the effectiveness of aerosolising our lentiviral (LV) vector carrying the reporter gene LacZ, using a vibrating mesh nebuliser (Aeroneb®Pro) (Figure1). It has been speculated that the virus particles on the surface of aerosols are subjected to surface tension and shear stress[1]. The aim of this study was to compare to test effectiveness of liquid vs aerosol delivery of LV-LacZ and to see if efficacy could be improved by suspending the virus in a range of diluents.

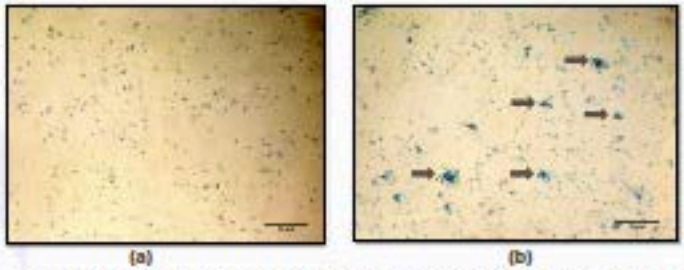
Figure1: Aeroneb®Pro used to aerosolize LV-LacZ

METHODS:



RESULTS:

- The distribution of the gene expression produced by the aerosol was generally homogenous across the culture (Figure 2a) compared to the clusters observed (arrows on Figure 2b) when using the liquid bolus delivery.
- The transduction obtained via aerosol was 33% to 51% of the number of cells compared to liquid control, for 1:250 to 1:1000 dilutions of the virus (Figure 3).
- Levels of transduction were lower with aerosol delivery compared to liquid delivery when virus was suspended in different diluents (Figure 4).
- Virus suspended in MS+BSA showed significantly higher levels of transduction when compared to virus suspended in PBS (Figure 4).



(a) (b)

Figure 2: (a) Aerosol delivery of LV-LacZ vector using the Aeroneb® Pro (1:250 dilution), (b) Liquid bolus delivery of LV-LacZ vector (1:250 dilution)

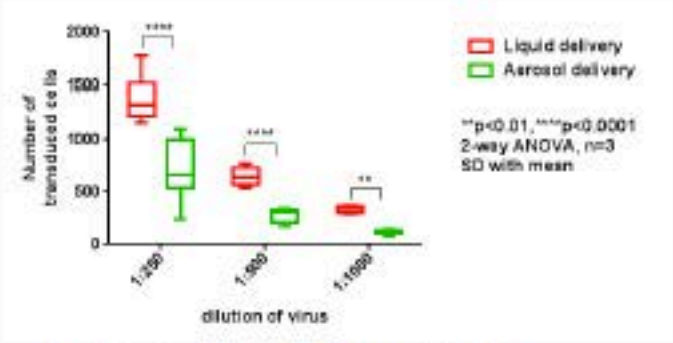


Figure 3: Comparison of liquid vs Aerosol delivery of LV-LacZ vector

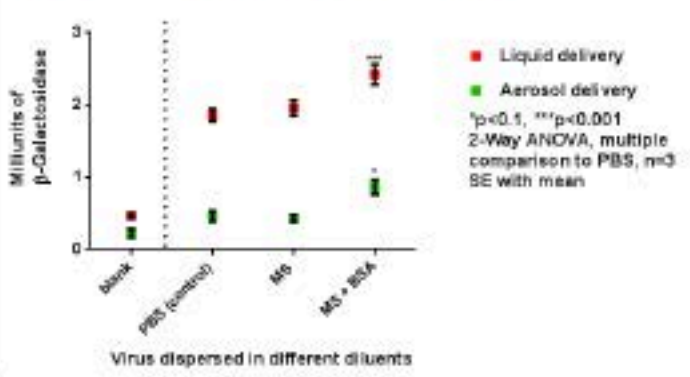


Figure 4: Testing the effect of different protective agents in which the virus is suspended

CONCLUSION:

- CHO cells can be transduced by an LV aerosol delivered using a vibrating mesh nebuliser (Aeroneb®Pro).
- Although the levels of transduction were lower in the aerosol group than the liquid bolus group, the dispersion produced by the Aeroneb®Pro may be advantageous in improving transduction of the conducting airways *in-vivo*.
- We speculate that the presence of MS+BSA aids in protecting the LV from shear stress.
- To improve the levels of gene transduction we plan to test other potentially protective agents and different nebulization platforms.
- These findings assist in our understanding of LV aerosolisation characteristics and provide practical information for future testing into the lungs of animal models and ultimately for CF airway disease.

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1. Yang W, Marr LC. Mechanisms by Which Ambient Humidity May Affect Viruses in Aerosols. Appl Environ Microb. 2012;78(19):6781-8.