Lentiviral airway gene therapy correction of CFTR function in knockout cystic fibrosis rats

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INTRODUCTION

- Cystic Fibrosis (CF) is a life limiting genetic disease, resulting from mutations in CF transmembrane conductance regulator (*CFTR*) gene.
- CF results abnormal ion transport in the airway epithelium, with lung disease being the main cause of morbidity and mortality in those that have CF.
- Airway gene therapy is considered one way to be a key method to prevent or treat lung disease in all patients with CF.
- Therapeutic efficiency of airway gene transfer can be quantified by perfusing the nasal membrane with specific salt solutions and measuring the ion transport, known as nasal potential difference (PD).
- A *CFTR* knockout (KO) rat shows altered consistent with the CF profile.

AIM

The aim was to assess the therapeutic benefit of lentiviral CFTR gene vector delivery to the nasal epithelium of CF KO rats.

METHODS

- Nasal PD were first optimised in wild-type (WT) and CF KO rats.
- The right nostril of CF KO adult rats (n = 16) were conditioned with 5 µl of 0.3% LPC, followed one hour later by 20 µl of LV-*CFTR* vector with a V5 epitope tag (LV-V5-*CFTR*).
- Prior to, and one week following LV delivery, CFTR function was assessed using nasal PD measurements.

RESULTS

Ion channel activity can be restored in the nasal epithelium of CF KO rats after gene addition treatment with LV-V5-CFTR

- Seven days after delivery of LV-V5-*CFTR* the nasal PD response in CF KO rats was towards WT response levels, shown by figure 1 and 2.

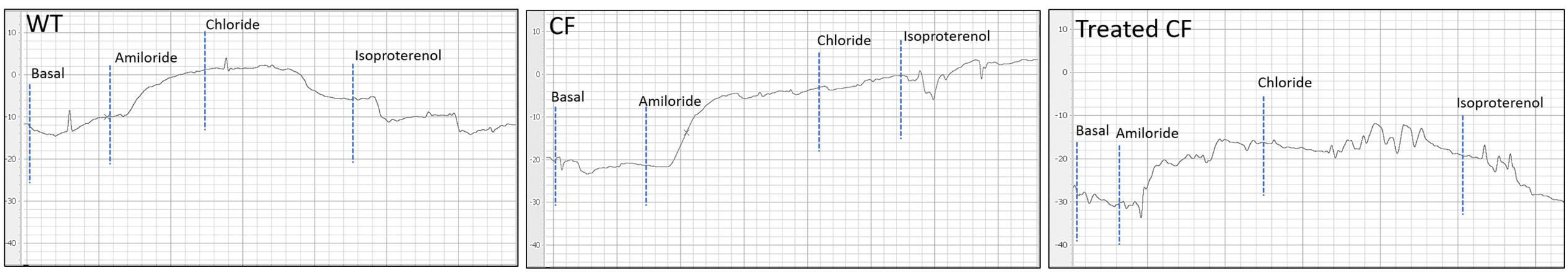

Figure 1: Representative nasal PD racing of WT, CF KO and treated CF KO rat.
When compared to WT, CF KO rats demonstrate classic CF electrophysiological defects in nasal respiratory epithelium. LV-V5-*CFTR* treated CF KO rats show correction of the bioelectrical defect.

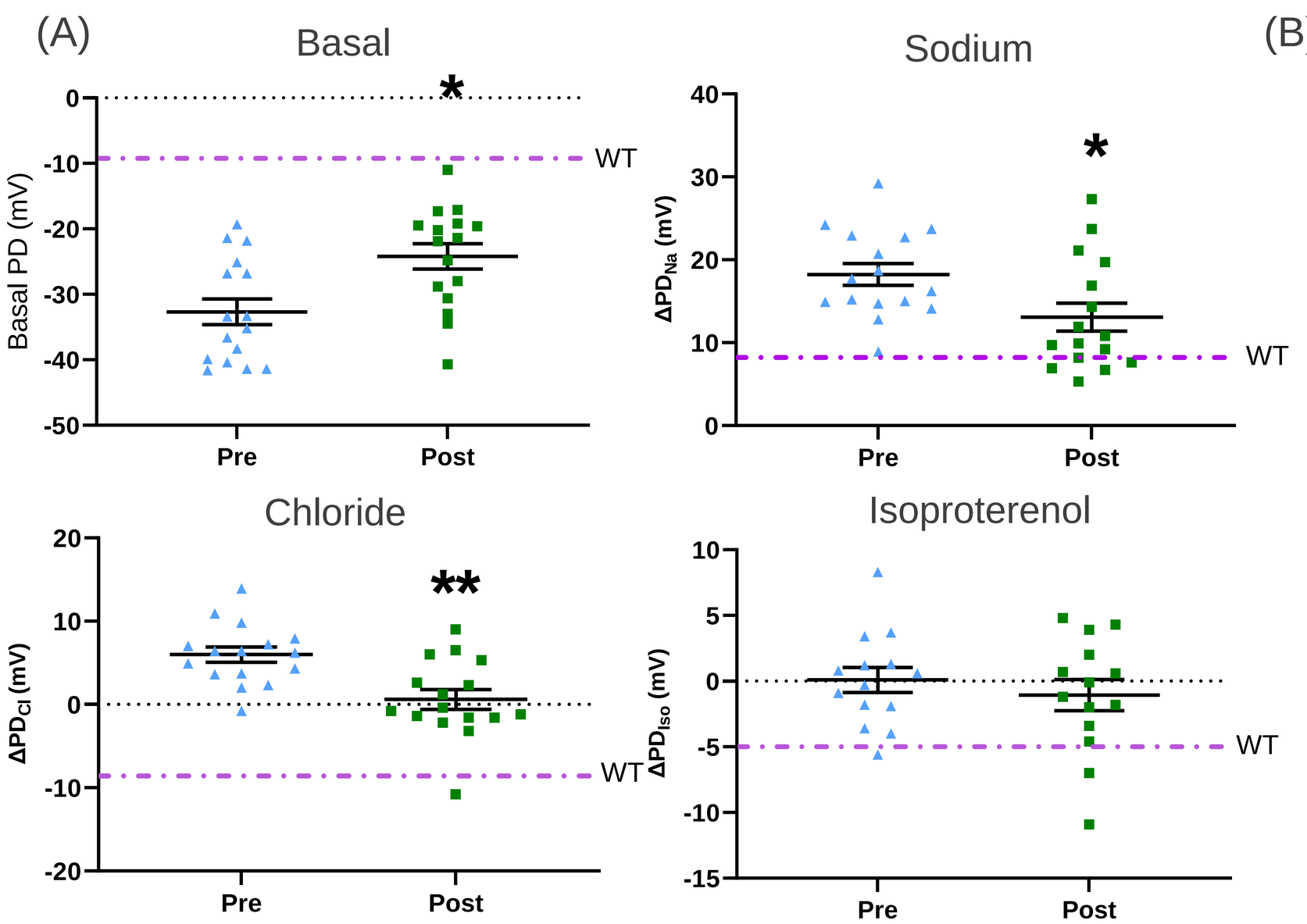

Figure 2: Pre-treatment and post-treatment nasal PD results of LV-V5-*CFTR* vector delivered to nasal epithelium in CF KO rats.

(A) Nasal PD measurements in CF rats for basal KRB, and ΔPD_{Na}, ΔPD_{Cl} and ΔPD_{Iso}. Seven days after delivery of LV-V5-*CFTR* CF rats low chloride response was significantly different to the pre-treatment response, with correction of 46% towards WT level. Data represented as the mean with SEM. Dot/dash line indicates the WT mean. (* $p \leq 0.05$,** $p \leq 0.01$ paired t-test; n = 16).

(B) Immunohistological detection of V5 tag (red) in the rat nasal epithelium confirms the success of *CFTR* gene addition in CF KO rats.

CONCLUSION

Using optimised PD methods we have shown successful correction of the bioelectrical defect in CF KO rats for the first time, after LV-V5-CFTR delivery.

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