




Treatment of cystic fibrosis airway cells with CFTR modulators reverses aberrant mucus properties *via* hydration

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CFTR rescue failed to normalise acidic pH, but reduced mucin concentration. Extended rehydration, not pH adjustment, facilitated mucus removal from cell surfaces, suggesting that mucus hydration is the dominant biochemical change in CF airways. <https://bit.ly/3iKUzFd>

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Abstract

Question Cystic fibrosis (CF) is characterised by the accumulation of viscous adherent mucus in the lungs. While several hypotheses invoke a direct relationship with cystic fibrosis transmembrane conductance regulator (CFTR) dysfunction (*i.e.* acidic airway surface liquid (ASL) pH, low bicarbonate (HCO_3^-) concentration, airway dehydration), the dominant biochemical alteration of CF mucus remains unknown.

Materials/methods We characterised a novel cell line (CFTR-KO Calu3 cells) and the responses of human bronchial epithelial (HBE) cells from subjects with G551D or F508del mutations to ivacaftor and elexacaftor-tezacaftor-ivacaftor. A spectrum of assays such as short-circuit currents, quantitative PCR, ASL pH, Western blotting, light scattering/refractometry (size-exclusion chromatography with inline multi-angle light scattering), scanning electron microscopy, percentage solids and particle tracking were performed to determine the impact of CFTR function on mucus properties.

Results Loss of CFTR function in Calu3 cells resulted in ASL pH acidification and mucus hyperconcentration (dehydration). Modulation of CFTR in CF HBE cells did not affect ASL pH or mucin mRNA expression, but decreased mucus concentration, relaxed mucus network ultrastructure and improved mucus transport. In contrast with modulator-treated cells, a large fraction of airway mucins remained attached to naïve CF cells following short apical washes, as revealed by the use of reducing agents to remove residual mucus from the cell surfaces. Extended hydration, but not buffers alkalised with sodium hydroxide or HCO_3^- , normalised mucus recovery to modulator-treated cell levels.

Conclusion These results indicate that airway dehydration, not acidic pH and/or low $[\text{HCO}_3^-]$, is responsible for abnormal mucus properties in CF airways and CFTR modulation predominantly restores normal mucin entanglement.