Suppression and Reversal of Cigarette Smoke-Induced Inflammasome Activation/Activity and Lung Injury by Novel Mitochondria-Targeted Sulfide Delivery Molecules

M. Whiteman[1], D. Pacitti[1], A. Brown[2], R. Torregrossa[1], L. Balachandran[1], V. Kumar[2], N. Hansbro[3], M. E. Wood[4], T. J. Haw[2], C. Scotton[1], P. M. Hansbro[3]; [1]University of Exeter Medical School, Exeter, United Kingdom, [2]Hunter Medical Research Institute, University of Newcastle, Newcastle, Australia, [3]Centenary Institute, University of Technology Sydney, Sydney, Australia, [4]Biosciences, University of Exeter, Exeter, United Kingdom.

Corresponding author's email: m.whiteman@exeter.ac.uk

RATIONALE: The major risk factor in the development of chronic obstructive pulmonary disease (COPD) is cigarette smoke (CS) with ~ 50% of smokers developing the disease. Thus, interventions to prevent/reverse CS-induced lung damage offer a novel treatment option in COPD and related lung diseases. CS has very recently been shown to deplete lung levels of the newly identified endogenous anti-inflammatory signalling molecule, hydrogen sulfide (H_2S). Lung/sputum H_2S levels negatively correlate with smoking pack years, are depleted in COPD, and further lowered after CS exposure. During oxidative stress (such as occurs in the COPD lung, or after CS exposure) H_2S-synthesising enzymes translocate to mitochondria and H_2S stimulates ATP synthesis, inducing cytoprotective mechanisms and indicating mitochondrial H_2S is beneficial to the lung in regulating cellular bioenergetics. Pharmacological inhibition or genetic silencing of endogenous H_2S synthesising enzymes exacerbates lung inflammation, mitochondrial damage and oxidative injury indicating that impairment of H_2S synthesis and/or loss of H_2S bioavailability is detrimental in COPD and negatively impacts on mitochondrial health. METHODS: We have investigated whether novel slow release selective mitochondria-targeted H_2S delivery molecules (mtH_2SD) we have developed (e.g. AP39 and RT01) could suppress and/or reverse CS-induced inflammation and lung injury. To examine the effects of mtH_2SD on suppressing COPD development, female C57Bl/6 mice were exposed to CS (12 cigarettes, b.i.d, 5 days/week) or air for 8 weeks with (1.0 mg/kg i.p.,q.d.). The effects of mtH_2SD on reversing CS-induced lung inflammation and remodelling were also assessed: mice were exposed to CS for 8 weeks as described above followed by either 4 weeks of rest or continued CS exposure, each with mtH_2SD (1.0 mg/kg i.n.,q.d.). Airway inflammation was assessed by differential enumeration of inflammatory cells in bronchoalveolar lavage fluid (BALF) and BALF IL-1β levels determined by ELISA. Lung function was assessed by forced oscillation and forced manoeuvre techniques. RESULTS: CS exposure lowered lung H_2S levels and induced inflammasome activity. mtH_2SD significantly ($p<0.05$) suppressed CS-induced alveolar destruction, fibrosis and improved lung gas exchange (DF_{CO}). Administration of mtH_2SD to mice with experimental COPD (at 8 weeks), reversed CS-induced lung neutrophil, eosinophil and macrophage infiltration, loss of lung function, and partially reversed airway resistance in both

continued smoking and after rest models. CONCLUSIONS: Targeting H_2S to mitochondria may be a novel therapeutic approach to prevent and/or reverse mitochondria-driven inflammation and lung injury in COPD and related diseases.

This abstract is funded by: Medical Research Council (UK); National Health and Medical Research Council (Australia)