INFECTION EXPERIENCED LUNG STROMAL CELLS PROVIDE EARLY IMMUNE PROTECTION THAT IS INDEPENDENT OF THE ADAPTIVE IMMUNE RESPONSE

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Abstract S111 Figure 1 In obesity, antiviral immune responses are impaired and airway metabolic milieu is altered. (A) Bronchoalveolar lavage (BAL) cells from 15 obese and 15 non-obese control subjects were infected ex vivo with H1N1.09 (H1N1), seasonal H3N2 and B/Florida (B/Flo) influenza strains. (B) IFN-α, (C) IFN-β and (D) IFN-λ antiviral immune proteins at 24 hour post-infection. Quantification of leptin in obese vs non-obese subjects in (E) bronchosorption and (F) BAL. *p<0.05; **p<0.01; ***p<0.001; ns=non-significant.

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Methods C57BL/6 mice were infected intranasally with IAV (WSN) and after 30 days re-challenged with IAV (X31). Mice were sacrificed at day 0, 2, 30 and 32 post-infection/re-infection. The transcriptional profiles and locations of infection experienced stromal cells were identified by RNAscope, viral nucleoprotein (IAV-NP) was detected by immunofluorescence and flow cytometry. An in vitro co-culture assay measured T cell re-activation by naïve or IAV infected stromal cells. To assess the consequences of T cell depletion on viral load following IAV re-challenge, anti-CD4/CD8 blockade was performed during the memory phase of infection.

Results IAV infection led to formation of dense clusters of T and B cells. RNAscope revealed high expression of SpiB and Cxcl0 in lung epithelial cells and fibroblasts, specifically at sites near immune cell clusters. SpiB regulates genes involved in antigen processing/presentation, while Cxcl10 facilitates T cell communication, suggesting collaboration between these cell types could facilitate early viral control. However, depletion of CD4 and CD8 T cells prior to a re-challenge infection did not alter enhanced viral control in IAV-memory animals. To investigate whether IAV-memory stromal cells display enhanced intrinsic viral control, we infected stromal cells from naïve and IAV-memory mice with IAV in vitro. Less IAV-Nucleoprotein was found in stromal cells taken from memory than naïve animals, this was independent of type I interferon. Interestingly, the NP+epithelial cells in cultures of IAV-memory stromal cells expressed higher levels of MHCII than NP+epithelial cells from naïve animals. Both populations could present IAV antigens to CD4 and CD8 T cells, although infected IAV-memory stromal cells to a lesser extent than infected naïve-stromal cells, perhaps due to lower levels of antigen.

Conclusions IAV-experienced lung stromal cells can play dual roles in anti-viral control, early cell intrinsic control and a rapid ability to communicate with local T cells.

CELLULAR SENESCENCE AMELIORATES HUMAN RHINOVIRUS CLEARANCE

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Abstract S113 Figure 1 In obesity, antiviral immune responses are impaired and airway metabolic milieu is altered. (A) Bronchoalveolar lavage (BAL) cells from 15 obese and 15 non-obese control subjects were infected ex vivo with H1N1.09 (H1N1), seasonal H3N2 and B/Florida (B/Flo) influenza strains. (B) IFN-α, (C) IFN-β and (D) IFN-λ antiviral immune proteins at 24 hour post-infection. Quantification of leptin in obese vs non-obese subjects in (E) bronchosorption and (F) BAL. *p<0.05; **p<0.01; ***p<0.001; ns=non-significant.

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Background Accumulation of senescent cells is one of the key hallmarks of ageing, and its senescence-associated secretory phenotype (SASP) has been shown to cause chronic inflammation, which contributes to age-related diseases. Aged populations are more vulnerable to respiratory viral infections and are likely to develop more severe complications after infections, especially the aged individuals with underlying disease such as chronic obstructive pulmonary disease (COPD). However, aging (cellular senescence) effects on respiratory viral infections was not demonstrated in the

Aim To investigate the role of cellular senescence on human rhinovirus (HRV) infections.

Method Cellular senescence was induced in human foetal lung fibroblast cells MRC-5 cells by a treatment of Nutlin 3A, a p53 stabilizer, and in human air-liquid interface (ALI) cultured induced pluripotent stem cells (iPSCs)-derived bronchial epithelium (HiLung Inc.) by a treatment of etoposide (2µM) for 2 days. Both cell types demonstrated elevated p21, a marker of cellular senescence. The cells were inoculated with HRV16, and viral load and CXCL8 in supernatants were determined by a 50% tissue culture infectious dose (TCID₅₀) assay and ELISA, respectively.

Results A peak viral load of HRV16 on Day 2 post inoculation was not different between senescent or normal MRC5 cells, but the viral load at later time point (Day 4 post inoculation) was higher in senescent MRC5 cells, suggesting delayed virus clearance compared with non-senescent MRC5 cells. When the senescent cells were treated with a senolytic combination cocktail, Dasatinib + Quercetin, virus clearance delayed in senescent cells was restored. In addition, viral replication was more rapid in etoposide treated senescent ALI iPSCs bronchial epithelium compared to non-senescent cells, and higher levels of pro-inflammatory cytokine CXCL8 release were also observed in senescent iPSC bronchial epithelium.

Conclusion This study demonstrates that senescent status affects HRV replication or elimination, supporting the age-dependent susceptibility to HRV infections. Selective elimination of senescent cells by a senolytic agent will be a promising option for preventing severe complications of infections from occurring in the elderly.

Question: Is using FeNO in primary care practicable and effective in asthma management?

Design Multi-centre study with cluster randomisation. Subjects recruited from Thames Valley GP practices. Subjects: new asthma and poorly controlled asthmatics. Day 0: full assessment symptoms, medication, history and FeNO. Trial visits: day 30 and 60 for assessment and correction of inhaler technique and adherence. FeNO driven treatment algorithm was followed vs standard care. Final visit d360. Goal: maintain FeNO <25 ppb. Primary outcome: ACT score. Data for the first 71 participants from 251 total.

Results Poorly controlled cohort: Fifty-five adults completed trial using FeNO guidance. 76% non-smoking, mean age 56 years, with mild to moderate airflow obstruction. 87% were using ICS and 58% reported good asthma control at baseline (ACT >19). Baseline and D360 data shown in table 1. A FeNO of <25ppb was achieved and maintained in 54%, compared baseline, p<0.001. There was a modest increase in ICS dose from 400 mg to 800 mg daily, no change in exacerbation rate (p=0.823). ACT score improved from median 20 (16,23) at baseline, to 21 (19,24), p=0.020 at 12 months. FeNO levels were lower at study end, median 24ppb vs 28ppb at baseline (p=0.085).

New Asthma Cohort: 16 enrolled, 11 completed, 63% female, mean age 40yrs. At study end ACT and MRC scores were improved, mean ICS dose: 400 mg Bec. FeNO was unchanged.

Conclusion: This initial analysis demonstrates that asthmatics in primary care improve control when FeNO guides ICS dose adjustments This was both practicable and acceptable in the primary care setting. However, continued clinical support is required for continued improvement.

'S115 EFFICACY OF HIGH-DOSE TRIPLE THERAPY ON ASTHMA EXACERBATIONS IN ASTHMATICS WITH PERSISTENT AIRFLOW LIMITATION AND HIGH BLOOD EOSINOPHIL COUNT: A POST-HOC ANALYSIS OF THE TRIGGER STUDY

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Background High-dose triple therapy with ICS/LABA/LAMA is recommended for adults with uncontrolled asthma in GINA Steps 4–5 and is often administered concomitant with or prior to initiation of biologic treatment. Previously, we reported in a post-hoc analysis of two large randomized clinical trials (TRIMARAN & TRIGGER) that patients with asthma uncontrolled on ICS/LABA and exhibiting persistent airflow limitation (PAL) may particularly benefit from the addition of LAMA (Singh D et al. Eur Respir J 2020). Here we explore the efficacy of high-dose ICS triple therapy in patients not controlled by high-dose ICS/LABA exhibiting both PAL and high blood eosinophil count, a phenotype that is considered for a step-up to biologic therapy.

Methods Using the dataset from the TRIGGER study, we conducted a post-hoc analysis in subjects with asthma uncontrolled by high dose ICS/LABA exhibiting PAL (post-salbutamol FEV₁≤80% and FEV₁/FVC≤0.7) and a blood