lower ER accumulation of polymeric-F-AT ($P < 0.001$). In the F-AT cell, PERK mRNA was upregulated at 3h compared to 0.5h in Z-AT. Although elevated compared to M-AT cells, F-AT cells had lower NF- B activity ($P < 0.001$), TNF-a production ($P = 0.046$) and IL-6 production ($P = 0.012$) compared to Z-AT.

In conclusion, F-AT secretion was comparable to M-AT. However, secreted F-AT was defective as an anti-elastase. F-AT was found to polymerise and aggregate in inclusion bodies. ER accumulation of F-AT activated the ER overload response; PERK-dependant-NF- B mediated inflammatory response, greater than M-AT but to a lesser degree than Z-AT. This data indicate that FZ phenotype may be at risk for liver and lung disease.

S64 CIRCULATING POLYMERS ARE FOUND IN ALPHA-1-ANTITRYPSIN DEFICIENCY AND ARE ASSOCIATED WITH LUNG DISEASE

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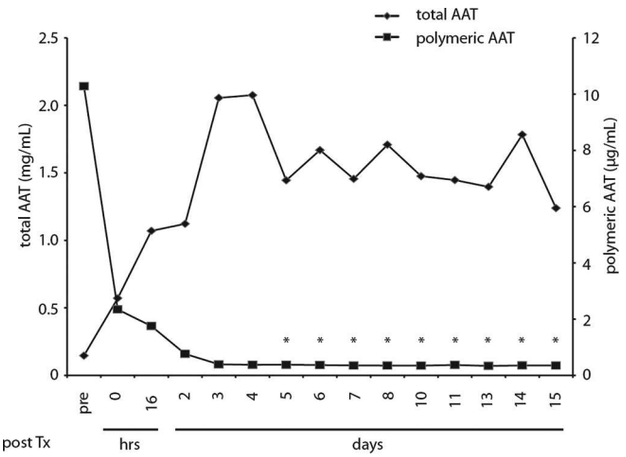
10.1136/thoraxjnl-2013-204457.71

Introduction and Objectives The severe Z deficiency allele of alpha-1-antitrypsin ($Glu^{342}Lys$) results in the formation of polymers that are retained within hepatocytes, leading to hepatitis, cirrhosis and hepatocellular carcinoma. The concomitant lack of circulating protein predisposes to early onset emphysema. Polymers are also found in the lung, skin and kidney and are known to be proinflammatory, but it is unknown whether those polymers are produced locally or are deposited from a circulating source. We wished to establish whether polymers are present in the plasma of individuals with alpha-1-antitrypsin deficiency, from where they originate and whether they are associated with any clinical phenotype.

Methods We used a novel anti-alpha-1-antitrypsin polymer monoclonal antibody (2C1) in an ELISA assay to evaluate whether polymers are present in a cohort of 513 individuals with ZZ alpha-1-antitrypsin deficiency. Serial samples from an individual with ZZ alpha-1-antitrypsin deficiency undergoing liver transplantation were used to investigate the source of circulating polymers. We then used a 2^{nd} cohort of 293 individuals with mixed alpha-1-antitrypsin phenotypes to determine whether circulating polymers could be used as a screening test for the presence of a polymerogenic allele. Disease associations were sought using clinical data from the ZZ alpha-1-antitrypsin deficient cohort.

Results In the cohort of 513 individuals with PiZZ alpha-1-antitrypsin deficiency, we found 512 had quantifiable polymers present within serum, the 513^{th} having previously had a liver transplant. Circulating polymer levels were higher in men and individuals with COPD and a there was a correlation with older age and lower lung function. The presence of circulating polymers was 100% sensitive and 89% specific in identifying 20 PiZZ alpha-1-antitrypsin homozygotes in a mix of 293 alpha-1-antitrypsin genotypes. Serial blood samples from a PiZZ individual undergoing liver transplantation showed that circulating polymers originate from the liver, clearing with a half-life of approximately 30 hours and becoming undetectable 5 days after transplantation.

Conclusions We have shown that circulating polymers are present in PiZZ alpha-1-antitrypsin individuals and originate from the liver. Polymer levels are associated with COPD, suggesting they may play a role in disease pathogenesis.



Abstract S64 Figure 1. Z 1 antitrypsin polymers are present in the circulation and are cleared following liver transplantation. Mouse monoclonal antibodies that detect all conformers of 1-antitrypsin, or only polymeric (1-antitrypsin (2C1), were used to quantify total (1-antitrypsin and (1-antitrypsin polymers respectively using sandwich ELISA. Time points marked * are below the lower limit of quantification (0.4 ug/mL).

S65 LARGE AND SMALL AIRWAY EPITHELIAL CELL SENESCENCE PRESENT IN COPD AND BRONCHIECTASIS?

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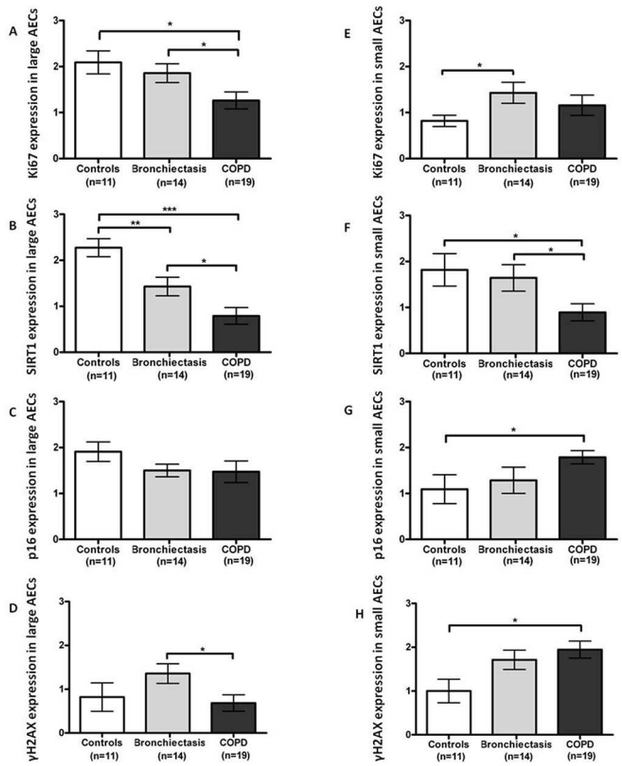
10.1136/thoraxjnl-2013-204457.72

Introduction and Objectives Accelerated lung ageing has been implicated in the pathogenesis of COPD and therefore targeting cellular senescence may have therapeutic benefit. COPD is increasingly felt to have significant sub-phenotypes with large and small airway involvement. The airway epithelium likely endures the majority of potentially senescence-inducing insults. However, data on airway epithelial cell (AEC) senescence in COPD is limited and comparisons between large and small airways are lacking. Furthermore, the role of infection in COPD-associated senescence is unclear. To date, senescence in bronchiectasis has not been investigated as a model for infection-induced senescence. We sought to determine AEC expression of senescence-associated markers in COPD and bronchiectasis and to compare large and small airways.

Methods Lung explant tissue from our transplant programme from COPD ($n = 19$) and bronchiectasis ($n = 14$) with resection tissue from smokers without lung disease (control) ($n = 11$) was stained for senescence-associated markers by immunohistochemistry. Staining was quantified semi-quantitatively. Fluorescence *in situ* hybridisation (FISH) was used to investigate telomere length and possible co-localisation with DNA damage-associated proteins.



Abstract S65 Figure 1. Expression of Ki67 (A), SIRT1 (B), p16 (C) and H2AX (D) in large AECs and small AECs (E, F, G and H, respectively). Results are expressed as mean ± SEM. Statistics: Kruskal-Wallis and Mann-Whitney U test. * p < 0.05, ** p < 0.01, *** p < 0.001.

Results In large AECs, expression of the proliferation marker ki67 and the anti-ageing protein sirtuin 1 (SIRT1) was decreased in COPD as compared to bronchiectasis and controls. There was no difference in expression of the cell-cycle inhibitor p16 and the DNA damage associated foci ?H2AX between the three groups. In small AECs, SIRT1 was decreased in COPD compared to controls and p16 and ?H2AX were increased. Here, ki67 expression did not differ between groups. In bronchiectasis, there was no significant change in senescence marker expression compared to controls, with the exception of decreased SIRT1 in large AECs. Marker expression was not significantly correlated with FEV_1 or smoking history. Preliminary work suggests potential co-localisation of ?H2AX at telomeres with ongoing analysis underway.

Conclusions Differential expression of senescence-associated markers between large and small airways in COPD may reflect the distinct patterns of inflammation and functional impairment occurring in the two airway compartments. There is some evidence suggesting a role for senescence in bronchiectasis, though this is less clear than for COPD. Further markers need to be investigated.

S66 TARGETING ANTI-AGEING MOLECULE AMPK RESTORES CORTICOSTEROID SENSITIVITY IN COPD

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10.1136/thoraxjnl-2013-204457.73

Rationale Chronic obstructive pulmonary disease (COPD) is an irreversible inflammatory lung disease and is currently the fourth greatest burden of disease of worldwide. However a key issue is that patients show a lack of response to corticosteroid treatment. Corticosteroid insensitivity is mainly caused by oxidative stress which directly stimulates inflammatory transcription factors and reduces the activity of co-adaptor proteins essential for the inhibitory actions of corticosteroids. AMP-activated kinase (AMPK) is a serine/threonine protein kinase that regulates cellular energy homeostasis and anti-oxidant defences, and has recently been labelled as an anti-ageing molecule. We hypothesised that activation of AMPK using Quercetin reverses corticosteroid resistance caused by cigarette smoke extract (CSE) in a monocytic cell line.

Methods Human monocytic cell line, U937s, were initially incubated with Quercetin (20μM) for 24 hours and then exposed to CSE for 2 hours. Cells were then treated with dexamethasone ($1x10^{-11}$ to $1x10^{-6}$M) for 45 minutes and stimulated with TNF-a (10ng/ml) for 24 hours. Supernatants were collected and CXCL-8 was measured using ELISA. Corticosteroid resistance was calculated as the ability of dexamethasone to inhibit 25% of TNF-a-induced CXCL-8 (IC_{25}). Activation of AMPK by Quercetin was measured using the levels of the phosphorylated AMPK by Western Blot. Nuclear factor erythroid related factor 2 (Nrf-2) levels and glucocorticoid receptor (GR) nuclear translocation were also assessed using Western Blot.

Results CSE induced corticosteroid resistance in U937s (IC_{25} = 30nM vs IC_{25} = 5nM). Interestingly Quercetin restored corticosteroid sensitivity by approximately 3 fold (IC_{25} = 11nM) compared to CSE. Quercetin increased levels of activated AMPK and also up-regulated the expression of Nrf-2. However, Quercetin was unable to restore GR nuclear translocation.

Conclusions Quercetin was found to be a potential novel therapy for restoration of corticosteroid sensitivity in COPD. Although the mechanism of action remains to be elucidated, Nrf-2 and AMPK activations which increase anti-oxidant levels and prevents oxidative damage could be a key the mechanism of action Activation of AMPK could therefore be a potential novel mechanism for the restoration of corticosteroid sensitivity and Quercetin could be used as an add-on treatment to corticosteroids in COPD.

Delivering better, safer care

S67 COPD–IN THE NEWS!

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10.1136/thoraxjnl-2013-204457.74

NICE[1] recommends 'track and trigger' systems for all adult acute hospital admissions and the RCP[2] advocates the use of the National Early Warning Score (NEWS) to promptly highlight high risk patients. In non-selected medical patients a higher admission NEWS correlates with higher mortality, with a stepwise increase as the score increases. Anecdotally patients with COPD have high rates of NEWS alerting. No studies have looked at the validity of the NEWS in COPD patients though the issue was raised in a previous abstract.

We retrospectively interrogated an electronic observation database in our Trust (two acute sites) over a year (February 2012–January 2013). We compared acute medical unit (AMU) admissions aged over 50 years (n = 13,291) with patients admitted with a primary diagnosis of COPD (n = 1119). Despite a similar age profile (median 74 & 77) and inpatient mortality