interventional trials. Participants in this CF study had a field-based estimate of exercise capacity and objective measurement of physical activity at each study visit.

Methods Participants wore a pedometer for 7 days prior to each visit. At each visit, London patients performed a standard incremental shuttle-walk test and Edinburgh patients a modified shuttle test (in which running was allowed). Data are expressed as mean (SD).

Results Data were analysed from 192 patients over 648 visits. Age at enrolment was 24 (11.9) years (London) and 20.8 (9.9) (Edinburgh) (p=0.052); FEV_1 was 67 (17.7)% and 79 (19.5)% for each site respectively (p<0.001). Daily step count at visit 1 was 7491 (2887) in London and 8872 (4089) in Edinburgh (p=0.04) and this difference persisted across subsequent visits. The coefficient of variation (CV) in step counts between visits was 21.3%. Number of shuttles completed in London was 61 (15), and Edinburgh 90 (33) with no trend over the four visits (CV=10 and 16% respectively). In Edinburgh there was a correlation between mean step count and the number of completed shuttles (r=0.46, p<0.001). Step count from both sites, and the number of shuttles completed in Edinburgh, correlated with FEV_1 % predicted (r=0.24, p<0.001 and r=0.27, p<0.001 respectively) and with age (r=−0.28, p<0.001 and r=−0.30, p<0.001 respectively). Such correlations were either weaker or not observed in London, however, in this group, number of shuttles correlated with height (r=0.51, p<0.001).

Conclusions No changes were detected in exercise capacity or daily activity levels over time. Between site differences were observed in both measures; however, these populations also differ in age and FEV_1. The modified shuttle test performed in Edinburgh appeared to better correlate with clinical markers than the standard incremental shuttle test performed in London, and is independent of height. We believe that testing exercise capacity is important in CF and we plan to investigate the other testing methods in the run up to our Multi Dose Gene Therapy Trial.

Funding Funded by the UK CF Trust.

COPD: sputum and exacerbations

P110 QUANTITATIVE PCR-BASED DETECTION AND QUANTIFICATION OF ATYPICAL BACTERIA AT BASELINE AND EXACERBATION OF COPD

doi:10.1136/thx.2010.150987.11

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Introduction Airway bacterial infections are associated with exacerbations of COPD. The potential role of atypical bacteria as a trigger for exacerbations is not well understood. Atypical bacteria such as *Chlamydophila pneumoniae* (CP), *Legionella pneumophila* (LP) and *Mycoplasma pneumoniae* (MP) are difficult to culture as they are intracellular pathogens. LP can be detected by urinary antigen, and serology can be performed for MP, but these techniques give no indication as to the atypical bacterial load. Quantitative PCR (qPCR) offers an alternative approach to identification and quantification of bacteria in sputum.

Methods Multiplex qPCR was used to detect and quantify CP, LP and MP in 238 samples prospectively collected from 87 patients in the London COPD Cohort: mean (±SD) age 71.4 (±8.1); predicted FEV_1 43.4% (±17.5%); male gender 47.9%; current smoker 49.2%. Baseline (n=104), exacerbation (n=95), and follow-up (n=39) samples were tested: Baseline was defined as at least 6-weeks without exacerbation, and exacerbation was defined as 2 consecutive days of two symptoms (Anthonisen criteria), at least one of which is a major symptom (dyspnoea; sputum purulence; sputum volume). Follow-up involved taking samples 2 or 5 weeks post-exacerbation onset. Using a qPCR developed by our clinical diagnostic service, the CP, MP and LP gene targets were RNA-polymerase β-chain; P1 adhesin protein; and MIP respectively. Routine microbiological analysis was also performed on these samples.

Results No samples were positive for the atypical organisms using culture. With qPCR analysis 6/238 samples (six separate patients) were positive for LP (2.5%), four at baseline and two at exacerbation/follow-up. One baseline sample was positive for MP (0.42%), and no samples were positive for CP. Atypical bacteria were present at 0.83% of exacerbations. Median (IQR) bacterial load was $4.3x10^4$ cfu ml^{-1} ($2.0x10^4$–$8.55x10^4$) for LP PCR-positive samples; the MP-positive sample load was $2.64x10^7$ cfu ml^{-1}.

Conclusion Quantitative PCR was more sensitive and informative than standard microbiological culture for the detection of atypical bacteria. Atypical bacteria in sputum were detected at very few exacerbations of COPD; moreover, when they were detected by qPCR, the load was low, indicating little or no significance in the aetiology of these events.

P111 FTIR SPECTROSCOPIC PROFILING OF COPD SPUTUM: IDENTIFICATION OF DISTINCT SPECTRAL SIGNATURES AND CORRELATION TO COPD STATUS

doi:10.1136/thx.2010.150987.12

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COPD remains common and complex with huge costs on the NHS and disruption to patients' daily lives. This presents a challenge for an effective, non-invasive test to enable determination and monitoring of COPD status. Fourier transform infrared (FTIR) spectroscopy for sputum profiling is timely. FTIR identifies and measures chemical bond vibrations within functional groups in complex biological mixtures by producing infrared absorption spectra. Identifying COPD-relevant spectra in sputum could provide sensitive rapid information on status and exacerbations. To address this, we randomly recruited 102 patients independent of severity. Sputum was collected at initial visit and in each of subsequent 2 weeks. Dyspnoea and sputum scores were obtained; FEV_1, serum CRP and exhaled NO measured; any intervening chest infection or treatment change documented. Patients was stratified by FEV_1; 26 patients had mild COPD ($FEV_1 \geq 80\%$); 41 moderate (FEV_1 49–79%); 35 severe ($FEV_1 \leq 50\%$). FTIR was performed using an Alpha-T spectrometer (Bruker UK); transmission mode in 4000 to 900 cm^{-1} region; 4 cm^{-1} resolution. All COPD sputa gave reproducible biological IR spectra with distinct signatures in 5 key regions at 3300–3280 cm^{-1} (assigned as amide A), 3000–2800 cm^{-1}, 1660–1600 cm^{-1} (Amide I), 1560–1520 cm^{-1} (Amide II) and 1180–1000 cm^{-1} (glycoproteins). Multivariate analysis showed significant correlation between spectral profiles and FEV_1 and smoking habit. The accuracy of differentiating mild from severe COPD was consistently greater than 70% (AUC under ROC curve); specifically we observed peak shifts in Amide A towards 3300 cm^{-1} as COPD worsened (p<0.001). Also 68/102 patients exhibited a clear band around 2060 cm^{-1}; the remaining 34 showed no band in this region. There was a significant association (p=0.012) between peak presence and COPD severity; with mild COPD patients more likely to have a peak, and severe sufferers having 2060 cm^{-1} signal absence. 35/102 patients had exacerbations during the study. Separate spectral analysis showed significant increase in glycoprotein max peak during COPD exacerbation (p<0.03). This study has important implications for future near-patient COPD monitoring. FTIR