Abstract S16 Table 1 Diagnostic performance of %VLFI and ODI >3% for detection of SDB in CHF patients

	%VLFI	ODI>3%
Sensitivity	0.53	0.97
Specificity	0.44	0.32
Positive predictive value	0.45	0.53
Negative predictive value	0.51	0.94
Positive likelihood ratio	0.94	1.42
Negative likelihood ratio	1.08	0.08
Area under receiver operating characteristic curve	0.49	0.92

Funding This study was funded by the British Heart Foundation.

S17 A PILOT STUDY OF THE PREVALENCE OF SLEEP DISORDERED BREATHING (SDB) AND NOCTURNAL HYPOXIA IN SYMPTOMATIC ADULTS WITH SICKLE CELL DISEASE (SCD) AND ITS RELATIONSHIP WITH DISEASE SEVERITY

doi:10.1136/thx.2010.150912.17

[1]P Murphy, [2]R Dillon, [3]A J Williams, [2]J Howard, [1]N Hart. *[1]Guy's & St Thomas' NHS Foundation Trust and Kings College London NIHR Biomedical Research Centre, London, UK; [2]Department of Haematology, Guy's & St Thomas' NHS Foundation Trust, London, UK; [3]Lane Fox Respiratory Unit, Guy's & St Thomas' NHS Foundation Trust, London, UK*

Introduction There are few effective therapies available for the long-term management of the cardiac and renal sequelae of SCD. Identifying reversible factors, which exacerbate disease severity, would facilitate development of new therapies or novel applications of established treatments. Nocturnal hypoxia (NH) merits investigation as a disease modulating factor as it is established that hypoxia promotes polymerisation of sickle haemoglobin and this is reversible with oxygen therapy (Noguchi *et al*, 1993). Although NH is common in children with SCD and is associated with poor outcome, similar data for adults with SCD are lacking. This is the first study to determine the prevalence of OSA and NH and quantify the severity of NH in adults with SCD. In addition, we investigated the correlation between the degree of NH and organ dysfunction.

Method Patients attending SCD clinic had an Epworth sleepiness score performed. Patients with either an ESS ≥10 or symptoms suggestive of SDB were offered nocturnal oximetry. Nocturnal oximetry findings were objectively scored and compared with the detailed clinical datasets collected at regular clinic attendances. OSA was defined as 4% oxygen desaturation index (4% ODI) of >10 events/h and NH was defined as >30% total sleep time (TST) with SpO_2 <90%.

Results 93 patients were screened. 34 had ESS ≥10 or clinical symptoms suggestive of SDB. 22 underwent nocturnal oximetry; mean ESS 12±4, clinic SpO_2 96±4%, 4% ODI 8±6 events/h, nocturnal SpO_2 91±4%, %TST SpO_2 <90% 43±41%. Prevalence of OSA and NH was 59%. The degree of nocturnal hypoxia was correlated with urine protein:creatinine (r=−0.35, p=0.02), elevated pulmonary artery systolic pressure (r=−0.71; p=0.0001) and prevalence of priapism (p=0.004). There was no difference detected in frequency of painful crises or hospital admission in patients with significant NH compared to those without NH.

Conclusion This small pilot study showed that OSA and NH had a prevalence of 59% in symptomatic adult SCD patients. These data have demonstrated a correlation between the severity of nocturnal hypoxia and pulmonary hypertension, renal impairment and priapism. These observations have not previously been reported. The strength of these correlations could suggest a causal relationship, although this needs to be confirmed in a larger prospective trial. Future studies should investigate the relationship between OSA, nocturnal hypoxia and organ dysfunction and need to be focussed on interventions such as nocturnal oxygen and continuous positive airway pressure.

New assessments in cystic fibrosis

S18 LONGITUDINAL ASSESSMENT OF BIOMARKERS FOR CLINICAL TRIALS OF NOVEL THERAPEUTIC AGENTS: THE RUN-IN STUDY

doi:10.1136/thx.2010.150912.18

[1]E W F W Alton, [2]C Boyd, [3]S Cunningham, [1]J C Davies, [4]S C Hyde, [2]J A Innes, [4]D R Gill, [2]A Greening, [1]U Griesenbach, [1]T Higgins, [2]D J Porteous. *[1]Department of Gene Therapy, Imperial College, London, UK; [2]Western General Hospital, Edinburgh, UK; [3]The Royal Hospital for Sick Children, Edinburgh, UK; [4]Gene Medicine Research Group, Oxford University, Oxford, UK*

We will be undertaking a phase IIB clinical trial of repeated application of liposome-based gene therapy over a one year period in approximately 100 CF patients (Multidose Trial). In preparation for this, we sought to address two key questions. Firstly, could we define the optimal set of patients in which the therapy could both be delivered (good access to the airways via nebulisation), and in whom any therapeutic effect was measurable (one or more abnormal measures of lung disease). Secondly, in this set of 'can deliver—can measure' patients, which biomarker(s) could be powered to be the primary outcome measure for the trial. To address both questions, we undertook a study (Run-in), cross-sectionally assessing 'can deliver' and longitudinally assessing a large set of candidate biomarkers for 'can measure'.192 patients from age 10 upwards, with FEV_1 >40% were enrolled at two clinical centres; 154 of these remained in the study after four visits spaced at approximately 4–5 month intervals. Biomarkers assessed cross-sectionally included radionucleotide deposition scans, CT and mucocilary clearance. Longitudinal biomarkers included a large series of serum, sputum and exhaled breath inflammatory markers, lung physiology, exercise-related assays and quality of life assessment. 12 patients were judged too severe for adequate delivery and were excluded. A shortlist of 4 biomarkers was generated based on a) showing a CF/non-CF difference, b) response to course of intravenous antibiotics, and c) coefficients of variation. These four were matched against the remaining 142 patients, and a further seven patients excluded in whom none of these short listed biomarkers was abnormal. 89 patients (3 or 4 biomarkers abnormal) have been definitely included to progress into the Multidose Trial, and a further 46 (1 or 2 biomarkers abnormal) are awaiting the final primary outcome selection. The Run-in study has, therefore, been able to a) select a cohort of 'optimal' patients in which to assess gene therapy and b) provide an indication of which may be the more useful biomarkers to use in phase IIB clinical trials of novel therapeutic agents.

S19 REAL TIME PCR IN THE IDENTIFICATION AND MANAGEMENT OF *ASPERGILLUS* IN CF

doi:10.1136/thx.2010.150912.19

[1]C G Baxter, [2]A M Jones, [2]A K Webb, [1]D W Denning. *[1]University Hospital of South Manchester, Education and Research Department, Manchester, UK; [2]Manchester Adult CF Unit, Manchester, UK*

Purpose The reported prevalence of *Aspergillus fumigatus* in CF sputum varies widely from 12 to 57%. While patients with ABPA are routinely treated with antifungals, it is not know whether colonised

or sensitised patients would benefit from antifungal treatment. To aid treatment decisions and to monitor response more accurate methods to detect *Aspergillus* in sputum are needed. This study aimed to identify CF patients with *Aspergillus* colonisation, using real time PCR, and examine the relationship of colonisation to markers of sensitisation.

Methods 108 adult CF patients provided a sputum sample and a blood sample. Serological tests included total IgE, specific *A. fumigatus* IgE and specific *A. fumigatus* IgG performed by Phadia ImmunoCAP® assay, and *A. fumigatus* precipitins by counter immunoelectrophoresis. Sputum was homogenised with sputasol and sonication. 10 µl was cultured on sabouraud agar (Oxoid, UK) for 72 h. The remaining sample was used in a commercial real time PCR assay, MycAssay Aspergillus. Patients on antifungal treatment were excluded from serological data analysis.

Results 30% of the 108 sputum samples were positive for *Aspergillus* species by standard culture whereas 80% were positive for *Aspergillus* species by PCR. 15 patients were on antifungal therapy of whom 7 were PCR positive. Of the serological tests, only specific IgG correlated to positive PCR. Using a ROC curve, a specific IgG level above 65 mg/l gave 85% sensitivity and 100% specificity for positive PCR. 12 patients met the 2003 consensus minimum criteria for ABPA. All were PCR positive supporting the use of antifungals for ABPA. 38 patients were sensitised to *aspergillus* (specific IgE >0.4 KUa/l), 28 of these were PCR positive. A group of 32 patients was identified that had a rise in specific IgG and positive PCR but no IgE rise. They may represent '*aspergillus* bronchitis'. All patients with negative serology were PCR negative.

Conclusion Real time PCR can accurately identify CF patients with *Aspergillus* in their sputum, including those in whom antifungal therapy is inadequate. However, PCR alone cannot distinguish between ABPA, sensitisation and colonisation. Positive PCR correlates to a specific IgG >65 mg/l. A randomised trial of antifungal therapy is required to determine if there is clinical benefit in treating PCR positive patients.

S20 THE BACTERIAL CYTOSKELETON—A NEW ANTIMICROBIAL TARGET IN CYSTIC FIBROSIS PATHOGENS?

doi:10.1136/thx.2010.150912.20

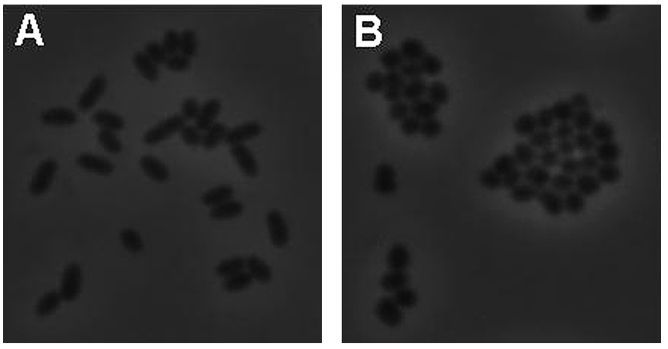
[1]S C Carnell, [2]J D Perry, [1]C M A Khan, [3]A De Soyza. *[1]Institute for Cell & Molecular Biosciences, University of Newcastle, Newcastle-upon-Tyne, UK; [2]Department of Microbiology, The Freeman Hospital, Newcastle-upon-Tyne, UK; [3]Institute of Cellular Medicine, University of Newcastle, Newcastle-upon-Tyne, UK*

Background *Burkholderia cepacia* complex (BCC) bacteria are opportunistic pathogens which cause severe lung infections in cystic fibrosis (CF) patients. Treatment of BCC infections is difficult due to the inherent multidrug resistance of BCC. There is a pressing need to find new bacterial targets for antimicrobials that provide functions essential for cell growth & replication. A major component of the bacterial cytoskeleton is the actin homologue MreB. MreB maintains bacterial cell shape by forming filaments under the bacterial inner membrane. A22 is a cell permeable compound that disrupts MreB, destabilising the bacterial cytoskeleton and altering the bacterial shape.

Aims To investigate the MreB bacterial cytoskeleton as a novel target for antimicrobials.

Methods We have tested a synthetic library of A22-related compounds and identified compound Q22 as a potential antimicrobial of interest against BCC and *Pseudomonas aeruginosa* strains. BCC bacteria have been grown in the presence of Q22 and a number of phenotypic changes observed.

Results Q22 inhibited growth of all 9 BCC species tested, including *B. cenocepacia*. A reduction in growth rate and cell morphology changes were also observed (Abstract S20 Figure 1). Higher concentrations of Q22 were required to exert *B. cenocepacia* growth effects (30 µg/ml Q22) when compared to *P. aeruginosa* (3 µg/ml Q22), probably due to the presence of two *mreB* genes in the *B. cenocepacia* genome. BCC bacteria lipopolysaccharide (LPS) is known to play an important role during infection. We analysed the LPS profile of BCC bacteria grown in the presence of Q22 and selected strains show profile differences when compared to untreated bacteria. The influence of Q22 treatment on bacterial motility and Type 3 secretion, a virulence associated secretion system, was assessed. However, growth inhibition masked motility analysis and differences observed in secreted protein profiles could not be attributed to Type 3 secretion. The growth conditions required for induction of Type 3 secretion *in vitro* remain undefined.



A, *B. cenocepacia* J2315 grown in LB with no additives;
B, *B. cenocepacia* J2315 grown in LB containing 30ug/ml Q22

Abstract S20 Figure 1

Conclusion *In vitro* MreB is an attractive new target for novel antimicrobials. Further analysis of current observations and additional phenotypic analysis will be required to dissect the nature of Q22-induced changes. Work supported by Newcastle-Upon-Tyne Hospitals Special Trustees and Italian CF Research Foundation (FFC).

S21 IDENTIFICATION OF *PSEUDOMONAS AERUGINOSA* INFECTION VIA VOLATILE ORGANIC COMPOUNDS IN SPUTUM HEADSPACE GASES

doi:10.1136/thx.2010.150912.21

[1]S Savelev, [2]J Perry, [3]S J Bourke, [1]R Taylor, [1]A J Fisher, [4]M Petrie, [1]P A Corris, [1]A De Soyza. *[1]Newcastle University, Institute Cellular Medicine, Newcastle upon Tyne, UK; [2]Dept Medical Microbiology Freeman Hospital, Newcastle upon Tyne, UK; [3]Cystic Fibrosis Unit, Royal Victoria Hospital, Newcastle upon Tyne, UK; [4]Newcastle University, Institute of Ageing & Health, Newcastle upon Tyne, UK*

Background *Pseudomonas aeruginosa* is a key respiratory pathogen with a distinctive odour in culture. An elevated level of hydrogen cyanide in the breath has been associated with the presence of *P. aeruginosa* in the airway, thus determining compounds specific to *P. aeruginosa* offers the possibility of a non-invasive diagnostic (breath) test.

Hypothesis Determining relevant to *P. aeruginosa* volatile compounds from sputum headspace gases offers target validation for the development of an electronic nose breath test for *P. aeruginosa*.