Prize symposium

**T3**

**HUMAN RHINOVIRUS IMPAIRS THE INNATE IMMUNE RESPONSE TO BACTERIA IN MONOCYTE DERIVED MACROPHAGES FROM PATIENTS WITH CHRONIC OBSTRUCTIVE PULMONARY DISEASE**

LI Finney, KBR Belchamber, P Mullia, SL Johnston, LE Donnelly, JA Wedzicha. National Heart and Lung Institute, Imperial College, London, UK

10.1136/thoraxjnl-2016-209333.3

**Introduction** Exacerbations of Chronic Obstructive Pulmonary Disease (COPD) are associated with accelerated disease progression, hospitalisation and death. Respiratory viruses are identified in approximately half of all exacerbations. We have previously found that rhinovirus infection leads to a secondary outgrowth of bacteria which is associated with increased exacerbation severity. The mechanisms of how HRV increases risk of secondary bacterial outgrowth are unknown.

**Hypothesis** We hypothesised that HRV infection impairs phagocytosis of bacteria by monocyte derived macrophages (MDM) which may lead to an increased risk of secondary bacterial outgrowth during COPD exacerbations.

**Methods** Participants were recruited from the London COPD Cohort. MDM were generated by culture in GM-CSF or M-CSF for 12 days. MDM were incubated with HRV 16 for 24 hours at increasing multiplicity of infection (MOI) 0.5–10 for 24 hours or poly-IC at increasing concentrations 0–300 μg/ml.

Phagocytic capacity was then assessed by incubating MDMs with fluorescently labelled heat killed *Haemophilus influenzae* or *Streptococcus pneumoniae* for 4 hours and uptake measured by fluorimetry.

The pro-inflammatory cytokine CXCL-8 and anti-inflammatory cytokine IL-10 were measured by ELISA according to the manufacturer’s instructions.

**Results** HRV16 impaired phagocytosis of *H. influenzae* (HRV (MOI of 5) 2.84 ± 0.92 vs media control 4.36 ± 1.06 RFU × 103 n = 8, p = 0.01) and *S. pneumoniae* (p < 0.01) by MDM in a virus-dose-dependent manner without impairing cell viability. HRV16 alone induced CXCL-8 and IL-10 release from MDM compared to media alone. HRV16 (MOI 5) significantly impaired IL-10 response to *H. influenzae* compared to media alone (0.59 (0.33–0.96) ng/ml vs 1.83 (1.11–3.00) ng/ml, n = 6, p = 0.03) and impaired CXCL-8 response to *H. influenzae* compared to media alone 4.41 (3.45–5.85) ng/ml vs 24.65 (11.63–29.77) ng/ml, n = 5, p = 0.01).

**Conclusion** HRV impairs phagocytosis of *H. influenzae* in a concentration-dependent manner without significantly impairing cell viability. Poly IC alone also induced IL-8 release from MDM.

**Conclusions** HRV impairs phagocytosis of bacteria by MDM in COPD and impairs cytokine response to bacteria which may inhibit neutrophil influx and prevent resolution of inflammation. This may lead to an outgrowth of bacteria and prolonged exacerbations in COPD.

---

**T4**

**GLOBAL SPREAD OF MYCOBACTERIUM ABSCESSUS CLONES AMONGST CYSTIC FIBROSIS PATIENT**

1DM Gregorano, 2JM Bryant, 3D Rodriguez-Rincon, 4Everall, 5KP Brown, 6P Moreno, 7D Verma, 8E Hill, 9Dijkkoning, 9CS Haworth, 10SR Harris, 12DOrdway, 12Parkhill, 1RA Floto, 1University of Cambridge Department of Medicine, Cambridge, UK; 2Wellcome Trust Sanger Institute, Hinxton, UK; 3Cambridge Centre for Lung Infection, Papworth Hospital, Papworth, UK; 4EMBL European Bioinformatics Institute, Hinxton, UK; 5Mycobacteria Research Laboratory, Department of Microbiology, Immunology and Pathology, Columbia University, Fort Collins, USA

10.1136/thoraxjnl-2016-209333.4

**Introduction** Lung infections with *Mycobacterium abscessus*, a species of multidrug resistant nontuberculous mycobacteria, have increased in frequency worldwide, emerging as an important global threat to individuals with cystic fibrosis (CF) where they cause accelerated inflammatory lung damage and death. *M. abscessus* was previously thought to be independently acquired by susceptible individuals from the environment. However, using whole genome sequencing and detailed epidemiological analysis of the majority of patients attending the CF centre at Papworth Hospital, we found strong evidence for transmission between patients. We therefore sought to examine the mechanism of acquisition of *M. abscessus* in CF individuals across the world.

**Methods** We undertook whole genome sequencing on 1080 isolates from 517 patients from the UK, US, the Republic of Ireland, mainland Europe and Australia. This was then correlated with clinical metadata and phenotypic functional analysis.

**Results** Our genomic analysis revealed that the majority of infections are from densely clustered *M. abscessus* genotypes with low levels of diversity, indicating a high level of human associated spread. Moreover, the phylogeny reveals the presence of three recently emerged dominant circulating clones that have globally spread. We found that these clones are associated with worse clinical outcomes and show increased virulence in both cell-based and mouse infection models. Within patients we found evidence of genetic diversity and evolutionary adaptation through the processes of convergent evolution and hypermutation.

**Conclusions** The majority of *M. abscessus* infections in patients with Cystic Fibrosis are caused by genetically related clusters, indicating recent patient-to-patient transmission despite conventional infection control measures. Transmission appears to have facilitated evolution of *M. abscessus* from an environmental organism into a transmissible human pathogen.

This work was supported by The Wellcome Trust grant 098051 (JMB, SH, JP) and 107032/A1 (DG, RAF) The Medical Research Council (JMB), The UK Cystic Fibrosis Trust (DMG, DR-R, IE, JP, RAF), Papworth Hospital (DMG, KPB, CSH, RAF), NIHR Cambridge Biomedical Research Centre (RAF), and The UKCRC Translational Infection Research Initiative (JP).