Results We have compared CF and non-CF bronchiectasis (n=36 combined). Polymicrobial communities were observed in all CF and non-CF bronchiectasis patients. However, CF patients demonstrated a greater bacterial diversity with a mean of 14.77 species per sample (range 6–21) than non-CF bronchiectasis patients who had a mean of 9.67 species per sample (range 4–14). However, fungal communities were similar between CF and non-CF bronchiectasis with 73.3% and 75% of patients harbouring fungi in their LRT respectively. Similarly, CF patients had a mean of 1.33 fungal species per sample (range 0–4) whilst non-CF bronchiectasis patients had a mean of 1.16 fungal species per sample (range 0–3).

Conclusions We note a complex microbiota in the lungs of both CF and non-CF bronchiectasis patients. In contrast to other studies using DNA based molecular analysis we note an increased microbial diversity observed in the CF cohort. The increases in bacterial taxa in CF may be due to differences in CFTR status, disease duration, or the intensive antibiotic regimens creating differing biological niches in non-CF bronchiectasis.

Paediatric infectious diseases

S24 DIFFERENTIATED PRIMARY BRONCHIAL EPITHELIAL CELL (PBECS), MONOCYTE DERIVED MACROPHAGES (MDMS) AND MONOCYTE DERIVED DENDRITIC CELLS (MODCS) TRANSWELL CO-CULTURE: RESPIRATORY SYNCYTIAL VIRUS (RSV) INFECTION OF THE APICAL AND BASOLATERAL SURFACES

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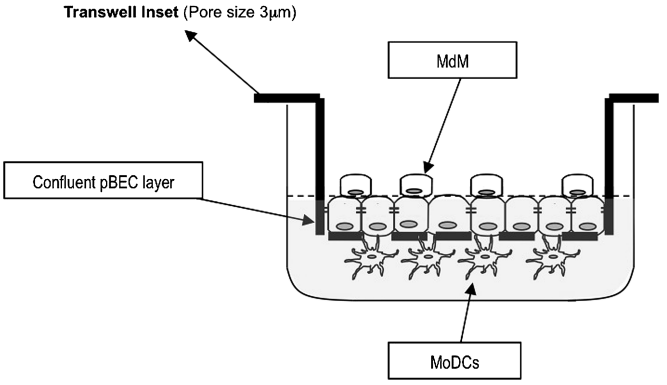
Introduction and Objectives RSV causes winter epidemics of respiratory disease. Active infection is virtually absent in summer months. Infected ciliated airway epithelial cells, local macrophages and dendritic cells secrete cytokines including interleukins (IL) 6 and 8, promoting a strong neutrophilic response that is important in disease clearance and airway inflammation. *In vitro*, RSV is capable of infecting MoDCs. RSV inhibits their maturation and can remain dormant in these cells. Dormant RSV can be stimulated to replicate with exogenous nitric oxide. These MoDCs are then able to re-infect HeLa cells (a lab strain of immortalised cervical cells). The following hypothesis was explored: *Infected dendritic cells act as a reservoir for RSV over summer months*.

Aim The aim of this study was to investigate the effect of RSV on pBEC and MoDc cell lines across a semi permeable membrane in the presence of MdMs.

Methods Primary bronchial epithelial cells were seeded in the apical part of the transwell model at 1×10^6 cells per ml and differentiated over 21 days on an air liquid interface. MoDCs were seeded on the basolateral part of the transwells at 1×10^6 cells/ml. MdM were seeded on top of the epithelial layer in selected experiments (see Abstract S24 Figure 1). Red fluorescent RSV (rr-RSV) was added to the apical side at a concentration of 1×10^6 plaque forming units (pfu)/ml with un-infected MoDCs on the basolateral side, and uninfected pBECs in the apical side were co-cultured with MoDCs previously infected with rr-RSV at 5×10^5 pfu/ml. Controls were uninfected pBECs with uninfected MoDCs or with just media on basolateral side. Red fluorescence (marker for active infection) was measured at 24, 48 and 168 h by flow cytometry.



Abstract S24 Figure 1

Results Directly exposed pBECs and MoDCs were infected at 24 h. Indirectly exposed pBECs were infected at 48 h. Indirectly exposed MoDCs were infected only when MdMs were present on the overlying epithelial cell layer.

Conclusions RSV is able to infect MoDCs and pBEC across a semi-permeable membrane in our *in vitro* model of airway epithelium, supporting MoDCs as a potential summer reservoir of RSV.

S25 IL 17 PRODUCTION IN PRIMARY AND SECONDARY RESPIRATORY SYNCYTIAL VIRUS (RSV) INFECTION AND NEUTROPHIL TRANSMIGRATION

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Introduction and Objectives RSV causes winter epidemics of respiratory disease particularly in infants and the immunocompromised. Infected ciliated airway epithelial cells, local macrophages and dendritic cells secrete cytokines including interleukins (IL) 6 and 8, promoting a strong neutrophilic response that is important in disease clearance and airway inflammation. The IL 17 inflammatory pathway, which is important in surface immunity, has been little studied in RSV infection to date. The following hypothesis was explored: *The IL17 pathway is important in neutrophil chemotaxis and restriction of RSV replication*.

Methods A transwell model of the airway was devised, co-culturing A549s (an immortalised bronchial epithelial cell line) and neutrophils to study the effect of RSV and IL17 on the transmigration of neutrophils. Neutrophil transmigration was assessed using light microscopic immunocytochemistry. Nasal swabs were taken from infants with RSV positive Bronchiolitis (n=49), RSV negative Bronchiolitis (n=12), the symptomatic older siblings of RSV positive infants (n=15) and uninfected, asymptomatic children (n=20). Cytokines released (IL6/IL8/IL17a/IL21/TNF) were analysed by cytokine bead array.

Results IL 17 and RSV caused significantly increased number of neutrophils to transmigrate compared to either IL 17 or RSV alone. Primary RSV infections which caused hospitalisation, are characterised by high levels of nasal epithelial derived cytokines (IL 6, IL 8). Less severe secondary RSV infections are characterised by relatively low levels of IL 6 and IL 8 and relatively high levels of IL 17.

Conclusions There is evidence that IL 17 is synergistic with RSV induced IL 8 in promoting neutrophil transmigration *in vitro*. There is evidence to support IL 17 as an important cytokine restricting RSV production in secondary infection *in vivo*. This study suggests that the IL 17 pathway is important in the pathogenesis of RSV bronchiolitis and could be potentially important in the development of novel therapies for this ubiquitous and important cause of respiratory disease.