Impact of propidium monoazide treatment on CF bacterial community pyrosequencing analysis

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In the majority of cases, CF sufferers will die as a result of chronic bacterial lung infections. The need to maintain lung function in these patients means that characterising of these infections is vital. Recent molecular based studies have shown the level of bacterial diversity in CF sputum to be much higher than previously accepted. Understanding the CF lung in terms of its microbial ecology could benefit our understanding of disease progression and influence treatment regimens.

DNA derived signals can originate from both viable and non-viable cells. In order to draw ecological conclusions from community-based analyses of samples from the CF lung it is important to focus analysis on viable bacteria. To ensure only DNA from viable cells are analysed, samples can be treated with propidium monoazide (PMA), a membrane impermeable dye that interacts with extracellular DNA or DNA within non-viable cells, to prevent PCR amplification.

Here, the impact of PMA treatment on 16S rDNA pyrosequencing-based characterisation of CF lung microbial communities was assessed. Parallel community characterisation was performed on treated and untreated sputum sample aliquots from adult CF patients, with the resulting data compared statistically.

PMA treatment significantly impacts characterisation of bacterial communities in CF sputum samples when applied in conjunction with 16S rDNA pyrosequencing analysis, this may have significant impact on future treatment regimes.

The effect of social deprivation on Pseudomonas and staphylococcal colonisation in the UK cystic fibrosis population

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Background: Pseudomonas (PA) colonisation is an important risk factor for more severe disease progression in CF. Low socioeconomic status has been linked with poor outcomes in CF. We explored, for the first time in a UK-wide cohort, the longitudinal risks of PA and Staphylococcus (SA) colonisation and their relationship with socioeconomic status (SES).

Methods: We undertook a retrospective longitudinal cohort study of 4346 people with cystic fibrosis aged less than 20 years (21132 observations) in UK CF registry between 1995 and 2006. Census based indices of multiple deprivation (IMD) from the UK constituent countries were used as small area measures of SES. A generalized estimating equation approach was used to estimate the effect of covariates on the population odds of infection.

Results: People in the most deprived quintile have a 50% increased risk of PA colonisation at any time point compared to those in the least deprived quintile (adjusted OR 1.5 95%CI 1.2 to 1.8). Age, genotype and birth cohort were also significant in the final model for PA, but there was no evidence of a sex effect. People in the most deprived quintile have a lower risk of SA colonisation compared to those in the least deprived quintile (adjusted OR 0.8 95%CI 0.65 to 0.97).

Conclusions: Social deprivation is associated with an increased risk of Pseudomonas colonisation, but a lower risk of Staphylococcus colonisation in young people with cystic fibrosis in the UK. We speculate that patients from more deprived areas may spend more time in hospital and thus have greater exposure to other patients with PA.

We acknowledge the support of the MRC and the UK Cystic Fibrosis Trust.