

Pseudomonas Aeruginosa from CF Patients Form Larger Biofilms in Vitro Than Those from Subjects with Bronchiectasis

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Aims:

Chronic *Pseudomonas aeruginosa* (Pa) infection is commonly seen in chronic suppurative lung diseases. Pa forms biofilms in the cystic fibrosis (CF) airway enabling bacterial persistence. There is limited evidence for biofilm formation in non-CF bronchiectasis (Bx). We compared in vitro biofilm-forming phenotypes of Pa strains from Bx and CF patients and explored potential confounding by bacterial growth rates.

Methods:

Pa strains from airway samples of 70 CF and 70 Bx patients with chronic infection were inoculated at an optical density (OD₆₀₀) of 0.05 into a 96-well plate and a parallel culture tube before 6 hours of static incubation. Adherent biomass was stained with crystal violet (CV) in the 96-well plates, eluted by ethanol and absorbance at 550nm (A₅₅₀) measured; biofilm formation was defined as staining > 3SD above broth only wells. Parallel cultures were vortexed and growth measured as change in OD₆₀₀.

Results:

Proportionally more CF strains demonstrated biofilm formation (CF 81%; Bx 66%, Chi² p=0.03). In the group as a whole, strains from CF patients demonstrated a 1.7-fold greater CV staining than Bx strains (p=0.02). Change in OD₆₀₀ over 6 hours correlated with the 6 hour CV biomass assay confirming that bacterial growth contributes to this signal (Spearman r 0.64 (95% CI 0.53-0.73), p<0.0001 for all strains but increased CV staining in CF strains was present after correcting for growth (p=0.04).

Conclusion:

Biomass of Pa from CF airway cultures was increased when compared to similar strains from Bx patients using a conventional biofilm assay. This may reflect altered phenotypic adaptation in response to environmental differences between the CF and Bx lung. Care should be taken to consider bacterial growth as an important factor in biomass formation as it may confound data generated from this assay. An affiliated project to the CF Trust-funded Strategic Research Centre for *Pseudomonas* infection.