WS1.1 Evolutionary pathways in two dominant clinical *Pseudomonas aeruginosa* clones

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Objectives: *P. aeruginosa* constitutes a particular problem in CF patients due to its ability to persist in the airways. Two dominating clones with different genotypes, DK1 and DK2, have been found to infect many CF patients in the Copenhagen CF clinic. We investigated the adaptation and evolution *in patient* of these transmissible *P. aeruginosa* clones.
Methods: Since 1973 *P. aeruginosa* isolates from sputum of CF patients have been stored. These isolates represent bacterial evolution and adaptation covering up to 200,000 bacterial generations of growth in CF airways. We have combined full-genome sequencing of longitudinal isolates of *P. aeruginosa* from a number of chronically infected patients with global gene expression analysis and other types of phenotypic characterization.
Results: Genome sequencing showed that mutations in regulatory genes were frequent. Generation of AlgT regulated population diversity and colonization of different niches in the CF airways was found to constitute a specific route of adaptation for DK1. In contrast, generation of AlgT variants with conditional phenotypes and colonization of different niches in the CF airways was found to be the dominant platform for DK2 adaptation.
Conclusion: The two transmissible clones have followed different evolutionary trajectories, suggesting that there could be several routes towards persistence of colonization of the CF airways. In the DK1 clone several genetic alterations in regulatory genes have resulted in diverse populations within the same patient. In the DK2 clone a small number of early mutations in global regulatory genes fixed in the population seem to be important for the colonization success.

WS1.2 Three clinically distinct chronic pediatric airway infections share a common core microbiota

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Background: Sequence-based methods have detected many microbes in CF secretions that are not routinely cultured. Those microbes can be further classified as those that are common and abundant in CF (core species) and those that are rare and scarce (satellite species). This categorization provides a vital foundation for investigating disease pathogenesis and to improve therapy. However, whether the core microbiota of children with CF are unique or are shared with those of other lung infections is not well known.
Objective and Methods: Using 16S bacterial rRNA pyrosequencing and ecological statistical tools, we compared the core respiratory microbiota from three cohorts of children with distinct airway diseases: Protracted bacterial bronchitis, non-CF bronchiectasis, and CF. The subject groups also differed in age, geography, antibiotic and steroid use, culture results, and lung function.
Conclusion: Despite the above cohort differences, we found the average core respiratory microbiota of all three groups of children to be strikingly similar in diversity and species constituency, arguing against airway disease-specific microbiota during early infection. The shared core microbiota included both traditional pathogens and species not identified by standard laboratory culture methods. These results suggest that these different airway infections share common origins, and that disease-specific microbes are not selected at an early stage. Each infection likely begins as a result of impaired clearance of the same airway microbiota, and early infection treatments successful for one of these diseases may be beneficial in the others, and perhaps for many other chronic lung infections.

WS1.3 Respiratory microbiota dynamics in newborns with cystic fibrosis and healthy controls: A longitudinal study

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Background: In Cystic Fibrosis (CF) patients, irreversible lung damage seems to begin in the first months of life, even if asymptomatic. Detailed colonization dynamics of the respiratory tract for this patient group have never been investigated this early in life.
Objective: To investigate the developing respiratory microbiota in CF infants and healthy age-matched controls in relation to respiratory health.
Methods: Case-control study cohort. Questionnaires and nasopharyngeal (NP) samples are collected at monthly to three-monthly intervals from CF patients and controls. In this preliminary analysis we studied the microbiota composition of 165 NP samples of 7 CF and 21 controls that were followed for at least 6 months, by 16SrRNA-based sequencing.
Results: Preliminary results show comparable bacterial density of respiratory microbiota in CF patients and controls, but higher diversity in controls at all time points. This coincided with a more stable colonization pattern over time in controls compared to CF infants. Predominant bacteria in both groups were Corynebacteria, Dolosigranulum, Staphylococcus, Moraxella and Streptococcus. In CF infants Corynebacterium, Dolosigranulum and Staphylococcus predominated in the first 3 months of life followed by Streptococcus and Moraxella. In controls Moraxella, Corynebacterium and Haemophilus predominated throughout the first 6 months of life.
Conclusion: We observed differences in bacterial colonization dynamics between CF and control infants especially in the first months of life. The influence of CF, antibiotic usage, and environmental exposure on microbiota composition and subsequent susceptibility to infections will be explored.

WS1.4 Significant bacterial infection missed using cough swabs compared to bronchoalveolar lavage in 1-year old newborn screened CF infants

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Introduction: Oropharyngeal cultures from cough swabs in children with CF are routinely used in clinical settings despite conflicting reports on their diagnostic accuracy [1,2]. While bronchoalveolar lavage (BAL) is an alternative diagnostic tool, BAL-directed therapy did not improve outcome in CF children at age 5 years [3].
Objective: To compare microbiological results obtained from BAL and oropharyngeal cough swabs at age 1-year in NBS CF infants.
Methods: As part of a multicentre observational study, elective bronchoscopic BAL sampled from at least 2 pulmonary lobes was obtained from asymptomatic NBS CF infants at 1 y. Corresponding cough swabs were obtained prior to the bronchoscopy.
Results: 39 BAL and swabs were obtained. Significant bacterial growth was found in 6/39 (15%) of BAL, (1 with *Pseudomonas aeruginosa*, 2 with *Staphylococcus aureus (SA)* and 3 with *Haemophilus influenzae*. Only 1 of these infections *(1 with SA)* was correctly detected by the swab with 2 further positive swabs not detected in BAL. Overall, swabs undertaken revealed only half of the infection (3/39; 8%) detected through BAL samples.
Conclusions: In NBS CF infants, vigilance and early treatment are likely to secure better long term outcomes but cough swabs predict lower airway infection poorly. While BAL may provide valuable information in specific clinical scenarios, better methods of detecting lower airway infection in NBS infants with CF are urgently required.

Reference(s)

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