Rnaseq As A Method To Study Microbial Interactions Arising In The Cystic Fibrosis Airways - DTU Orbit (08/11/2017)

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Introduction: In previous studies from our laboratory, a *Pseudomonas aeruginosa* lineage, named DK2, has been identified and characterized as highly successful, transmissible and persistent over four decades in cystic fibrosis (CF) patients. This lineage underwent substantial phenotypic and genetic changes over time and therefore provides a unique opportunity to explore the impact of those adaptational pathways on its ability to interact with other pathogenic bacteria such as *Staphylococcus aureus*, a pathogen frequently co-infecting the CF airways. Methods: We have used a novel method to study interspecies interactions between a CF isolate (2003) from the DK2 lineage and a wild-type *S. aureus* JE2. We grew both strains in mono or co-culture on LB agar, harvested RNA from the colonies after a 24-hour period. Subsequently we performed RNA-seq for the different samples. The data were then compared in a pairwise mode to isolate the transcriptomic profiles for each species. The most differentially expressed genes from both species were validated using real-time quantitative PCR. Results: Interestingly, the greatest expression change was observed in *S. aureus*, where large clusters of genes associated with virulence were differentially expressed, compared with the monoculture condition, while the *P. aeruginosa* DK2 response was much more discrete with isolated genes differentially regulated rather than whole operons or clusters. Conclusions: According to our data, *S. aureus* would display reduced virulence in the presence of an adapted *P. aeruginosa* DK2 clone, possibly as a consequence of the multiple hostile forces DK2 encountered over time during its long-term adaptation to the CF airways.

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