Synergistic interactions within polymicrobial biofilms formed by atypical and conventional species in cystic fibrosis

Susana Patrícia Peixoto Lopes (1), NF Azevedo (2), MO Pereira (1)
(1) University Of Minho, Portugal
(2) Faculty of Engineering, University of Porto, Portugal

A complex microbiome is present in Cystic fibrosis (CF)-airways, with uncommon species co-existing and establishing dynamic interactions with traditional pathogens. This study aimed to examine in vitro biofilm formation and susceptibility patterns of two CF-atypical bacteria, Inquilinus limosus (IL) and Dolosigranulum pigrum (DP), when associated to P. aeruginosa (PA), under oxygen-atmospheres resembling CF airways. The ability of IL and DP to form dual- and three-species biofilms with PA and to resist against antibiotics was evaluated under in vitro oxygen-restricted conditions. The fitness of biofilms was compared with monospecies consortia by individual species. Atypical species were able to develop biofilms with PA, presenting a great extent of adhesion in microtiter plate wells and achieving high cell-densities over 24h growth. Three-species populations were well-adapted to poor-oxygen environments, showing high growth rates. Both dual- and three-species biofilms were significantly resistant to most antibiotics, with minimum biofilm eradication concentrations in most cases not achievable (>1024 mg/L). Comparing with monospecies consortia, the presence of IL and DP did not disturb PA biofilms, resulting in most cases in polymicrobial biofilms with increased biomass, activity and antibiotic resistance, which persisted under oxygen-restricted environments. This study evidenced the contribution of some atypical species to develop resilient polymicrobial biofilms with PA in CF-airway oxygen conditions. Hence, CF treatment will only be successful after recognizing CF-infection as polymicrobial and upon exhaustive modulation of ecological processes, which will be useful to predict the effects of new therapeutic interventions. Authors acknowledge financial support from IBB-CEB, FCT and FEDER, trough Program COMPETE (project-PTDC/SAUSAP/113196/2009/FCOMP-01-0124-FEDER-01601), grant-SFRH/BD/47613/2008.

Frequent transcriptional switching of the cell wall adhesion gene, FLO11, mediates social behavior in yeast biofilm

Kristian Hanghoj (1), K Andersen (1), B Regenberg (1)
(1) Copenhagen University, Denmark

1. Introduction Phenotypic heterogeneity in biofilm opens the opportunity for social behavior between cells in the biofilm. In theory, social interactions occur if i) two phenotypes coexist and one or both gain a selective advantage, ii) resources are in surplus and iii) can create a selective advantage for genetically related individual. We propose that yeast biofilm exhibits a social behavior fulfilling these criteria through transcriptional switching of a cell surface adhesive gene. 2. Methods Wildtype Saccharomyces cerevisiae in the ∑1278b strain background switches expression of the FLO11 gene leading to mixed populations of Flo+ and Flo- cells in biofilm on semisolid complex medium. ∑1278b contain sfl1 had only Flo+ cells, flo11 had only Flo- cells. 3. Results We find that wildtype yeast biofilm populations containing both Flo+ and Flo- cells are larger than populations where all members are either Flo+ (5x) or Flo- (1.4x). This trait relies on frequent switching in FLO11 expression as shown by mutants depleted for factors responsible for the switch. We find that limitation of resources inhibits the selective advantage of transcriptional switching, as wt, Flo+, and Flo- colonies have similar population sizes when the population is prevented from migration. 4. Discussion Taken together, our results suggest that transcriptional switching of FLO11 is a social behavior in S. cerevisiae biofilm and that the interaction is only evident when migration takes place. Our data thereby fulfill the criteria for kin selection suggested by Hamilton (1967).