Correlations of *Flavobacterium columnare* colony morphotype with predator resistance, growth, biofilm-forming ability and virulence

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Abstract

The colony morphotype of the fish pathogen *Flavobacterium columnare* on agar plates can be rhizoid, rough or soft. Some strains can switch their colony type depending on e.g. nutrition, starvation and presence of phages. Colony type switching could serve as an important mechanism for *F. columnare* survival in the natural environment during free-living vs. infective phases, and may therefore correlate with other phenotypic changes. We investigated the correlations of virulence, resistance to protozoan predation, resource use, and biofilm-forming ability of 20 *F. columnare* isolates with rhizoid and rough colony morphotypes. The virulence of the rhizoid and rough colony phenotypes was studied using zebra fish infection model. Bacterial resistance to protozoan predation (amoeba *Acanthamoeba castellanii* and ciliate *Tetrahymena thermophila*), growth dynamics, and biofilm formation ability were studied in microcosm experiments.

We found that the colony type clearly correlated with the virulence of the bacteria: rhizoid type was significantly more virulent than rough type. In general, the tested *F. columnare* isolates were resistant to amoeba predation regardless of colony type, while the ciliates were able to consume bacterial biomass as food. Rhizoid and rough colony types showed different patterns in growth and biofilm-forming ability, but these patterns were specifically related to the isolate's identity rather than the colony type. Our results indicate that *F. columnare* isolates and their colony morphotypes may have different strategies to ensure their survival in natural environment. Our study is a first step towards understanding of complex trophic interactions between *F. columnare* and its predators outside the fish host.