Bioremediation and Sustainable Environmental Technologies—2017. Fourth International Symposium on Bioremediation and Sustainable Environmental Technologies (Miami, FL; May 2017). ©2017 Battelle Memorial Institute, Columbus, OH, www.battelle.org/biosymp. A. Barton and S. Rosansky (Chairs)

Bioaugmentation Using Engineered Polyvalent Bacteriophages

Jacques Mathieu (mathieu@rice.edu), Pingfeng Yu, Cory Schwarz, and Pedro J.J. Alvarez
(Rice University, Houston, TX, USA)

Background/Objectives. Bioaugmentation strategies have proven successful in the enhancement of biodegradation of targeted pollutants in a number of laboratory and field-scale trials. However, survival and activity of the exogenous microorganisms are not always achieved and are susceptible to failure for a variety of reasons. These include loss of catabolic plasmids, cell death due to incompatible environmental growth conditions (e.g., pH, DO, temperature, nutrient limitations, presence of inhibitory compounds, etc.), as well as competition by the indigenous microbial population and/or predation due to protozoan overgrowth. Strain selection is therefore considered an extremely critical factor for bioaugmentation. Gene bioaugmentation, which involves the introduction of catabolic genes into the environment via mobile genetic elements, has been proposed to circumvent the problems associated with cell survival. A basic premise of its application is that indigenous microbial soil species that uptake the introduced genes have already undergone strong selective pressures to optimize survivability in their ecological niche, so should be more fit than an exogenous species. Indeed, lab-scale studies have demonstrated the efficacy of gene bioaugmentation in several circumstances. However, gene uptake and transfer frequencies at pilot-scale are not always sufficient to facilitate contaminant catabolism. In an effort to address these limitations of bioaugmentation, we are developing technologies for isolating and engineering polyvalent bacteriophages to more promptly and effectively deliver catabolic genes of interest to indigenous microbial communities.

Approach/Activities. The extremely narrow host-range of most known phages limits their use to applications where prior knowledge of the microbial community structure exists. In contrast, extremely broad host-range (polyvalent) phages might be capable of circumventing this need. In this work, we report the successful isolation and characterization of polyvalent phages, and subsequently demonstrate their efficacy in both planktonic and biofilm environments of increasing host diversity. Genome sequencing of several polyvalent phages is reported, with the information being utilized to develop engineering strategies for catabolic gene expression.

Results/Lessons Learned. Our novel selection methods, which are optimized for use with multiple hosts, were capable of isolating widely polyvalent bacteriophages. In contrast to previous reports of polyvalent phages, these newly isolated phages displayed little reduction in efficiency on alternative hosts, even when infecting hosts of different phyla. Results to date indicate that polyvalent phages are particularly well-suited to complex, heterogeneous environments, such as biofilms, and are more effective for environmental use than narrow host-range phages. However, some polyvalent phages may present unique challenges for genome engineering. Nevertheless, our findings suggest that if broad dispersal of a catabolic gene or operon throughout a contaminated site is desired, then the use of broad host-range (polyvalent) phages may be warranted.