CONSIDER FOR TALK

10th Annual SEA Symposium Abstract

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The Mystery of Jamestown, Virginia (Phages)

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In an attempt to expand the diversity of Actinobacteriophage discovery, the 21 freshmen that comprised the William and Mary 2017-18 SEAPHAGES Lab attempted to identify phages that infected Corynebacterium glutamicum, and Tsukamurella paurometabola. After eight weeks and several hundred enrichment attempts no phage were isolated. Nearing November (and desperation) we turned to two Mycobacteria hosts: M. neoaurum and M. aichiense. Following over 50 enrichments, while no plaques were obtained from M. neoaurum, phage that infected M. aichiense were isolated by a single student and from the instructor’s “magic compost.” Despite apparent success with a novel host, in order to meet the sequencing deadline, William and Mary submitted Gorge, a phage isolated by our community college colleagues at the Colonial Williamsburg campus of Thomas Nelson Community College with whom we have collaborated for the past decade. Using M. smegmatis as a host, Gorge belongs to the F1 subcluster with high similarity to Saal. Concurrently with annotating Gorge, we continued to pursue our M. aichiense phage. Strikingly, in contrast to the rates of obtaining M. aichiense plaques from our SEAPHAGES class, in three separate high school outreach events during winter 2018, over half of the 66 high school students from Jamestown High school who conducted a phage discovery experiments at William and Mary obtained phage in their enrichment plates. These phage consistently show extremely low titers that decline quickly over time; moreover, even using three different methods of DNA extraction, we have not obtained significant amounts of DNA for sequencing although positive M. smegmatis and G. terrae lysates from previous years worked perfectly. For M. aichiense, all DNA was degraded following the DNAse step. We therefore performed shotgun cloning of non-DNAsed sample; while bacterial debris sequences comprised about 50% of the sequences, another percentage were phage sequences that showed some albeit low homology over very short stretches to F subcluster phages. These phages do not plaque on M. smegmatis. We are continuing to characterize these interesting and apparently novel phages along with the other 30 isolates from Jamestown, Virginia, using a variety of different approaches.