CONSIDER FOR TALK

8th Annual SEA-PHAGES Symposium Abstract

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Our Phage Phriends: Isolation & Characterization of Audrick & Porcelain

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As part of HHMI’s SEA-PHAGES (Science Education Alliance - Phage Hunters Advancing Genomics and Evolutionary Science) program, USciences students were able to isolate, characterize, and annotate two novel bacteriophages this year, Audrick and Porcelain. The phages both infected *Mycobacterium smegmatis* mc2155, but while Audrick was isolated using the enrichment protocol, Porcelain was isolated using a direct plating method. These phages were further characterized through the annotations of their genomes using DNAMaster, Starterator, Phamerator, and HHPred. It was revealed that Audrick is a C1 cluster, lytic phage that is part of the Myoviridae family with a genome length of 155,205 bp, and that Porcelain is a J cluster, lysogenic phage that is part of the Siphoviridae family with a genome length of 109,575 bp. Further observations with electron microscopy, and additional experiments such as purification through ultracentrifugation using a CsCl gradient, lysogeny tests, and cloning of a putative repressor were conducted to obtain a greater understanding of these phages and the similarities and differences between them. For Audrick, an interesting phenomenon that occurred during ultracentrifugation was the presence of three Schlieren lines; we expected only one band of extremely pure phage. After analyzing the different bands with electron microscopy, we visualized empty heads in the top band, intact phage particles in the middle band, and an unidentified substance speculated to be ribosomes in the bottom band. Images from the top and middle bands also showed evidence of tail activation similar to that observed in phage T4. For Porcelain, lysogeny tests were conducted using MiaZeal, a highly similar J cluster phage that was previously isolated and annotated at Cabrini College. Surprisingly, Porcelain infected MiaZeal’s lysogen. This result was intriguing because we had expected same-cluster phages to be homoimmune. Lastly, Porcelain’s putative CI repressor was successfully amplified through PCR and was transformed into *E. coli* MACH cells. Altogether, these results reveal novel and interesting phenomena related to each of these phage “phriends” that warrant further investigation.