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Isolation, Purification, and Genomic Analysis of the Novel Bacteriophage Kradal

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ISOLATION, PURIFICATION, AND GENOMIC ANALYSIS OF THE NOVEL BACTERIOPHAGE KRADAL Adeshola Fanegan, Katie Gemperli, Sofia Kling, and Jesse Yavner

Mentor: Chris Shaffer

Kradal, a novel bacteriophage isolated from the host Streptomyces griseofuscus, was isolated from a direct environmental sample collected just outside of Gaylord Music Library (38.6465 °N, 90.3112 °W) at Washington University in St. Louis, MO. Kradal was analyzed throughout five main phases: isolation, purification, amplification, extraction, and characterization via electron microscopy and gel electrophoresis, as part of the Science Education Alliance-Phage Hunters Advancing Genomics and Evolutionary Science (SEA-PHAGES) program sponsored by the Howard Hughes Medical Institute and Graham Hatfull at the University of Pittsburgh. Kradal produces uniformly circular, small, clear plaques when infecting Streptomyces griseofuscus. Additionally, electron microscopy classifies Kradal as a Siphoviridae phage with a prolate head measuring approximately 291 nm by 44 nm and a flexible tail with an average length of 280 nm. Through genetic sequencing using the Illumina Shotgun method, Kradal's genome length was determined to be 186,383 base pairs, which is the second longest sequenced phage genome to be sequenced in the SEA-PHAGES program. Kradal is closely related to the bacteriophage Satis, displaying similarity across 99% of the genome and in the same cluster, BM. Kradal's genome contains 335 protein coding genes, of which our group analyzed from 1-85 and from 254 to the end of the genome. Thirty-five of the genes in these sections of the genome run in the forward direction while 132 run in the reverse direction. Due to the novelty of Kradal, evidence from functional annotation and comparative gene analysis is limited. We are currently comparatively analyzing Kradal with Satis and various other Streptomyces phages with similar morphology and sequence to Kradal.