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ISOLATION AND GENOMIC CHARACTERIZATION OF STREPTOMYCES PHAGE CIRCINUS Alice Herrmann, Amy Kwan, Caelan Miller, and Brandon Tang

Mentors: Chris Shaffer and Kathy Hafer

A major goal of phage biology is to understand how the phage genome interacts with its bacterial host and how these interactions drive evolutionary change. Isolation and genomic characterization of environmental phages allows for this continuation of research into phage infection mechanisms. Streptomyces Phage Circinus was isolated from environmental sampling at the coordinates 38.6032 °N, 90.2616 °W near Seed Sprout Spoon in St. Louis, MO. Subsequent plaque assays and streaking with Streptomyces griseofuscus ensued to ensure presence of phage, with Circinus displaying clear, stellar plaques. Four rounds of purification and subsequent webbed plating were performed to achieve a high titer lysate of 6.0*109 pfu/mL. Transmission electron microscopy revealed that Circinus has an icosahedral head with a non-flexible tail with an average horizontal head, vertical head, and tail measurements of 85 nm, 80 nm, and 358 nm, respectively. Through genomic sequencing, Circinus was found to have a genome of 126,383 base pairs with a direct terminal repeat of 2050 base pairs and a G-C content of 52.9%. The draft annotation of Circinus contains 216 protein-coding genes with 33 tRNA genes. Circinus belongs to the BK cluster and BK2 sub cluster and shows a high level of similarity with the phage BillNye. Through meticulous manual annotation, our group found that the genome organization of Circinus is comprised predominantly of forward-strand genes, with a total of 10 reverse strand genes, split into a cluster of eight at 2620 bp to 6112 bp, with two scattered at 70279 and 119711 bp. By comparing similar proteins of previously annotated phages, we seek to discern the functions of the genes found in phage Circinus.