Characterization and Classification of a Montana Mycobacteriophage

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Froghopper, a Mycobacteriophage discovered by Nikki Boyd in 2005 and stored in Dr. Marisa Pedulla’s collection, was adopted in the fall of 2016. The bacteriophage was plated, or used to infect *Mycobacterium smegmatis* on Petri dishes, in order to determine the morphology of the resultant plaque. Froghopper was purified and amplified, and a high titer stock was made. DNA of the phage was extracted using phenol/chloroform. Restriction digests and agarose gel electrophoresis of Froghopper DNA were performed in order to compare the DNA of Froghopper to DNA of phages in the Actinobacteriophage database. The polymerase chain reaction (PCR) was used for preliminary determination of Froghopper’s phage cluster. A phage cluster is a group of bacteriophages with similar DNA sequences. Phage clusters can be predicted by a set of primers used in PCR to determine genetic similarities to sequenced bacteriophages (Smith et al., 2013). Determination of the bacteriophage’s structural morphology was determined by imaging the phage under transmission electron microscopy at the University of Montana. DNA of the bacteriophage was sent to the University of Pittsburgh for the DNA sequencing. Once sequenced, the DNA sequence was annotated; putative protein coding genes were identified and described in relation to other known sequences, and the annotated sequence was submitted to GenBank.