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The Story of Phage Gavriela: From a Moist, Dark Soil to Complete Genome Analysis

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Bacteriophages (phages) are viruses that strictly use bacterial hosts in their replication cycles. These viruses have practical uses vital to human and animal health, including phage therapy, genetic engineering, food safety, and vaccine development. Gavriela was isolated from a soil sample collected at LeTourneau University in Longview, Texas (32.468056° N, 94.725417° W), on August 27, 2024. The enrichment method was used with incubation at 37°C using Middlebrook 7H9 as the medium and *Mycobacterium smegmatis* mc2 155 as the bacterial host. A spot test was performed to confirm phage presence. 10-fold serial dilutions and three rounds of plating were used to purify the phage. Gavriela’s plaques looked cloudy and had an average diameter of 1.12 mm (range 0.6-1.8 mm; n =15). Phage Gavriela yielded a high titer lysate of 1.10 x1011 PFU/mL which was used for DNA extraction, TEM imaging preparation, and archiving. Gavriela displayed a siphovirus morphotype. Genome sequencing was done using Illumina NextSeq 1000 sequencing with ~1312 shotgun coverage. Annotation was done using various software and databases, including DNA Master, PhagesDB, NCBI, HHPred, Phamerator, Starterator, GeneMark, Glimmer, DeepTMHMM, ARAGORN, and tRNAscan-SE. Sequence data showed Gavriela to belong to subcluster P1 with a 13 bp 3’ sticky overhang (CCTGCCGCCCCGA), genome length 47,916 bp, and 67.3% GC content. 77 putative protein-coding genes (size range, 90 to 3651 bp) were predicted. Functions were assignable to only 52% (40/77) of the genes. Gabriela had the three key genes associated with a lysogenic life cycle, namely integrase, immunity repressor, and excise.