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Discovery and Genomic Characterization of Bacteriophage Xandras

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The bacteriophage population represents the largest reservoir of undiscovered genetic information in the biosphere. Here, we describe the isolation and genomic characterization of bacteriophage Xandras. Xandras was grown on *Mycobacterium smegmatis* and was enriched from a soil sample taken from a chicken coop in Beaver Dam, Kentucky. Xandras viral particles have a siphoviridae morphology with an average capsid diameter of 57 nm and an average tail length of 244 nm. It produces plaques with clear centers and turbid halos after 48 hours of growth at 30oC. The results of lysogeny experiments support the classification of Xandras as a temperate phage. Sequence analysis demonstrated that the genome of Xandras is 75,179 bp long with a 9 bp 3’ overhang of (CGCTTGTCA). BLAST analysis of the Xandras genomic sequence revealed that it belongs to the E cluster of mycobacteirophages and is most similar to mycobacteriophages StolenfromERC, Goldenspark and Americanbeauty. The E cluster of mycobacteriophages is composed of 127 members, with an average genome containing 75,488 bp, 142.6 genes, and 1.9 tRNAs. E-cluster phages are typically temperate and solely infect Mycobacterium hosts. Genome annotation of Xandras was completed using PECAAN (Phage Evidence Collection And Annotation Network), a program that collects evidence from a variety of resources to identify start and stop sites and predict functions of individual genes. Xandras is predicted to have 144 genes and 2 tRNAs. Many of genes have no known function (NKF) but potential conserved domains and/or potential functions were assigned to nearly 40% of the genes. The presence of a tyrosine integrase gene supports the observed plaque phenotype and provides a mechanism for lysogeny. Our results provide insight into the “viral dark matter” and provide additional evidence of the genetic diversity and mosaicism of mycobacteriophages.