A 2024 Report on the Characteristics of Phages Assigned to Subcluster AZ1

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Isolation and Morphological Characteristics

As of June 2024, 52 phages have been assigned to Actinobacteriophage subcluster AZ1. Subcluster AZ1 consists of temperate phages that were isolated from environmental samples collected from two countries (US and Canada) and 14 different US states, using *Arthrobacter globiformis and Arthrobacter atrocyaneus* as the hosts. A sampling of transmission electron micrographs reveals these phages to have siphoviral morphologies, while photographs of plaque assays reveal a range of morphologies, including turbid or clear and possible halos around the plaques. Plaques can be small (1-2mm) to large (4-10mm). Examples of phages displaying these morphologies include phages DrManhattan and ObiToo.

Genometrics

The genomes of phages in this subcluster have an average length of 43571 bp (range: 41,823 bp – 44,651 bp) with an average GC content of 67.2% (range: 65% – 68.8%). The genomes have 3’ sticky overhang ends.

Gene Content

a. A survey of all manually annotated genomes in this subcluster (i.e., excluding draft genomes) reveals the number of genes called per genome ranges between 65 – 74 genes, which represent a total of 171 gene phamilies (or phams).

b. Of these 171 phams, 35 are fully conserved across the subcluster. These phams are assigned putative functions that are involved in structure and assembly (e.g., terminase, portal protein, tape measure, tail assembly chaperones, and minor tail proteins), replication and recombination (e.g., DNA polymerase 1, RNA binding protein), and lysis (e.g., endolysin). Some other notable putative functions include Endonucleases VII, and G-I-Y-Y-I-G endonuclease.

c. Another 136 phams have varying levels of conservation. Notable here are SprT-like protease and serine integrase, which are conserved.

d. There are two different tRNA genes identified in this subcluster with one tRNA gene in four phages; thus, most of the AZ1 phages do not have any tRNAs. There is one identifiable

integrase but no identifiable repressor genes. This suggests that the phages of this subcluster do form lysogens and are, thus, temperate phages.

e. The genomes are organized with genes involved in structure and assembly located within the first third of the genome, while those involved in replication and recombination are scattered across the remaining two-thirds of the genome. AZ1 phages contain a single endolysin gene, but the location within the genome and the pham for the endolysin gene are inconsistent across the entire subcluster. At least three patterns are observed within the subcluster. Examples include Phives - Endolysin as gp1, Elezi - Endolysin in the middle of the genome (gp25), and Powerpuff - Endolysin in the right arm of the genome (gp59)

   Most genes are transcribed in the forward direction, with the exception of seven possible reverse genes. Two of the seven transcribed in the reserve direction are highly conserved genes. Most AZ1 phages have one or two reverse genes in their genome.

References

1. Klyczek KK, Jacobs-Sera D, Adair TL, Adams SD, Ball SL, Benjamin RC, Bonilla JA, Breitenberger CA, Daniels CJ, Gaffney BL, Harrison M, Hughes LE, King RA, Krukonis GP, Lopez AJ, Monsen-Collar K, Pizzorno MC, Rinehart CA; SEA-PHAGES Program; PHIRE Program; Staples AK, Stowe EL, Garlena RA, Russell DA, Cresawn SG, Pope WH, Hatfull GF. Complete Genome Sequences of 44 *Arthrobacter* Phages. Genome Announc. 2018 Feb 1;6(5): e01474-17. doi: 10.1128/genomeA.01474-17. PMID: 29437090; PMCID: PMC5794937.

2. Kapinos A, Aghamalian P, Capehart E, Alag A, Angel H, Briseno E, Corado Perez B, Farag E, Foster H, Hakim A, Hernandez-Casas D, Huang C, Lam D, Mendez M, Min A, Nguyen N, Omholt AL, Ortiz E, Saldivar LS, Shannon JA, Smith R, Sridhar MV, Ta A, Theophilus MC, Ngo R, Torres C, Reddi K, Freise AC, Moberg Parker J. Novel Cluster AZ Arthrobacter phages Powerpuff, Lego, and YesChef exhibit close functional relationships with Microbacterium phages. PLoS One. 2022 Jan 13;17(1): e0262556. doi: 10.1371/journal.pone.0262556. PMID: 35025964; PMCID: PMC8758107.

Source Data: https://observablehq.com/d/5e5bc78c9b3ae2ed accessed July 2024**.**

Acknowledgments
We thank Maddie Bendele, Isobel Cobb, and Steve Cresawn for developing the tools used to generate this report.