CONSIDER FOR TALK

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Mycobacteriophages Paphu and Philly: Two New Members of Two Familiar Clusters

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Sixteen new mycobacteriophages were isolated from soil samples collected around the state of Michigan and parts of the United States. All phages were capable of infecting Mycobacterium smegmatis and were isolated through either enrichment or direct plating at 32°C. A variety of plaque morphologies were produced based on size, shape, and clarity; both lytic and temperate phages appear represented in this collection. The mycobacteriophages, Paphu and Philly, were chosen as two of three phages for complete genome sequencing and comparative genomic analyses. The predominant plaque produced by Paphu after 48 hours at 32°C was circular and was approximately 2-3 mm in diameter. The plaque morphology was clear with a turbid ring resulting in a halo visual. The predominant plaque produced by Philly after 48 hours at 32°C was circular, clear, and was approximately 1 mm in diameter. The complete genome sequence for Paphu revealed a relationship to mycobacteriophages of cluster A, subcluster A1, which now contains 154 sequenced members. Paphu is most similar to the A1 phages AFIS and Blue. The genome size of Paphu is 51,159 bp, which is smaller than most of the sequenced A1 mycobacteriophages. However, it has one of the highest GC contents of A1 phages at 64.1%. The Paphu genome contains 92 protein-encoding genes and no tRNA or tmRNA genes. Despite the genomic identities organizing phages into subcluster A1, they show marked levels of genomic diversity, scattered throughout their genomes. The complete genome sequence for Philly revealed a relationship to mycobacteriophages of cluster B, subcluster B3, which now contains 31 sequenced members. Interestingly, all but a few B3 phages have been isolated between Michigan and Washington, D.C. The genome of Philly is 68,523 bp, which is smaller than most of the sequenced B3 mycobacteriophages. It has a GC content of 67.5%, similar to other B3 phages as well as the host M. smegmatis (67.4%). The Philly genome contains 102 protein-encoding genes and no tRNA or tmRNA genes. Phages in subcluster B3 share very high sequence identity throughout the genome lengths. Philly maintains this pattern, being nearly identical to mycobacteriophages Heathcliff, Athena, and Bernado despite their isolation in different years (2003-present) and in different geographical locations. However, Philly does contain some genomic variability around genes 2, 5, and 56. And like all B3 phages, Philly also contains many short sequence repeats throughout its genome.