CONSIDER FOR TALK

8th Annual SEA-PHAGES Symposium Abstract

Bucknell University

Lewisburg PA

Corresponding Faculty Member: Greg Krukonis (gregkrukonis@gmail.com)

The Arthrobacter phages of Bucknell University

Lisa Francomacaro , Lauren Otto, Christopher Bidlack , Kaitlyn Carduner , Sara Christian , Maxwell Everett , Paulina Gutkin, Ryan Kirby, Zachary Kozick, Madison Kremp , Elaney Marcotte , Eileen McAuley, Melville O’Brien, Samuel Sheridan , Nicholas Stamatos, Marie Pizzorno, Emily Stowe, Greg Krukonis

After isolating mostly small (15,524-15,630 bp) phages on Arthrobacter hosts (order Actinomycetales) belonging to cluster AN in 2013 and 2014, Bucknell made concerted efforts to isolate phages that would be more likely to belong to different clusters. We succeeded and isolated twelve novel phages, five of which were sequenced and annotated. All phages were isolated on Arthrobacter sp. ATCC 21022: Dino, cluster AK, 43,562 bp; LiSara, cluster AL, 60,637 bp; Tribby, cluster AM, 59,084 bp; Tophat, cluster AR, 70,091 bp, Teacup, cluster AU, 58,238 bp. These phages are (currently) the second or third phage annotated in their respective cluster.
Tribby and Teacup stand out as phages with a much lower GC content, 45.3 and 49.8% respectively, than the other three phages (61.1-64.7%), than many Actinobacteriophages, and than their Arthrobacter hosts (63.4% for ATCC 21022; Russell and Hatfull, 2016). The number of putative genes in these genomes ranged from 62 (Dino) to 111 (Tophat), with no tRNA genes in any genomes. Genes in these genomes are either all in the forward direction (Teacup) or have one small section (1 or 5 genes) of reverse genes. Transmission electro n microscopy revealed all isolated phages have a siphoviridae-like morphology.

Horizontal gene transfer is rampant in bacteriophages. To determine whether phages belonging to different clusters are part of similar or different networks of interacting phages and bacterial hosts, we blasted every putative gene product for our five sequenced phages against the tailed phages database on NCBI. For each gene, we recorded the genus of the bacterial host of phages with hits to that gene product. For example, for phage Tribby, genes 1 and 2 had blastp hits only to Arthrobacter phages, while gene 3 had blastp hits to Arthrobacter and Rhodococcus phages and gene 4 (an HNH endonuclease) had blastp hits to phages that attack bacteria belonging to 11 genera. For each phage, we then determine what proportion of its genes had blastp hits to phages of a particular genus. For example, for phage Tribby, 94.1% of its genes had blastp hits to proteins in other Arthrobacter phages and 2.9% to proteins in Rhodococcus phages, while for Teacup these percent are 90.7% and 40.7%, respectively. For our five phages, the percent of blastp matches to Mycobacterium phages ranged from 0% (Tophat), 1.6% (Dino), 14.7% (Tribby), 15.1% (Teacup), and 24.2% (LiSara). Given that we observed blastp matches to phages isolated on 150 genera of bacteria, Heatmaps were generated to compare the networks of these five phages. Given that only one phage was used for each cluster, further studies will need to include more members of each cluster to determine whether there are cluster-specific patterns.