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University of Maine, Honors College

Orono ME

Corresponding Faculty Member: Emily Whitaker (emily.whitaker@maine.edu)

ParABS encoding mycobacteriophage Polyphemus lacks an immunity repressor

Stanley Hollinger-Levitsky, Mason Clark, Ethan McEachern, Joseph Morse, Daniel Walsh, Emily E Whitaker

Many human bacterial pathogens, including non-tuberculosis mycobacteria, contain prophage-encoded virulence factors. Mycobacteriophage (phage), viruses which infect mycobacteria, can be maintained within the bacterial host as a prophage through integration into the host genome (lysogen formation), or as an extrachromosomal plasmid. Temperate phage carry out the lytic cycle and possess machinery necessary to form and maintain prophage while lytic phage cannot form prophage. Novel phage Polyphemus was isolated from garden soil collected in Edinburg, Maine using host *Mycobacterium smegmatis* mc2155. Polyphemus produces plaques ranging from 1 – 8 mm that feature a gradual increase of turbidity emanating from a clear center of the plaque. Polyphemus is a *Siphoviridae* family phage belonging to subcluster A6, with a 50,424-bp genome (GC content: 61.6%) that encodes 91 protein-coding and 3 tRNA genes. Polyphemus may utilize a relatively uncommon form of lysogeny maintenance. Polyphemus encodes a 1.4kb parABS cassette that could promote segregation of the Polyphemus genome as an extrachromosomal plasmid during cell division. However, attempts to isolate stable Polyphemus lysogens in *M. smegmatis* have not been successful. Intriguingly, the genome has a potential 2.3-kb INDEL event on the right arm where most A6 phage contain lysogeny maintenance genes, including an immunity repressor. Polyphemus is one of four phages that possess a parABS cassette while lacking an immunity repressor. Future studies should re-attempt lysogen isolation and superinfection immunity assays to clarify lysogenic potential. Additionally, the function of the parABS system within Polyphemus’ lifestyle should be further investigated for prophage establishment and maintenance.