Sequencing of Phage Isolates

Yersinia phages were isolated from raw sewage water harvested from the Livermore Water Reclamation Plant located in Livermore, California, USA. The host target strain for the LYP215 and LYP264 phages was Yersinia pestis CO2 pgm-. Y. pestis was grown using BHI at 28C. Ten mls. of sewage water were combined with 10 mls. of mid-log Y. pestis CO92 pgm- and incubated with shaking at 28C for 4 hours. The culture was centrifuged, and the supernatant was filtered through 0.2-um filter concentrator (Millipore, Billerica, MA, USA). The plaques were individually harvested and purified three times on the host strain. Phages were propagated using Y. pestis CO92 pgm- using double-overlay soft-agar method to generate a high titer lysate for generating DNA for sequencing.

Phage genomic DNA was extracted using the Genomic DNA Isolation Kit (Norgen Biotek Corp., Thorold, ON, Canada). Paired-end sequencing libraries were constructed using Nextera XT DNA library kit (Illumina, Inc., San Diego, USA). All libraries were quality checked using the Agilent BioAnalyzer (Agilent Technologies, Santa Clara, CA, USA). Sequencing was performed on Illumina MiSeq (Illumina, Inc., San Diego, CA, USA) using 2x300 v3 chemistry. For each sample dataset, the resulting sequence data was quality trimmed and assembled using CLC Genomics Workbench v.9.0.1 (Qiagen, Redwood City, CA, USA).