CONSIDER FOR TALK

8th Annual SEA-PHAGES Symposium Abstract

University of Texas at El Paso

El Paso TX

Corresponding Faculty Member: Germán Rosas-Acosta (grosas3@utep.edu)



Carlos Ontiveros



Jose Echeverri

Novel insight into the relationship between genomic sequence and bacteriophage morphology

Carlos Ontiveros, Jose Echeverri, Ana Alvidrez, Daniel Amparon, Kennedy Behrens, Robin Bonilla, Ibbel Bustillos, Giovanna Herrera, Maritza Hidrogo, Jesus Lujan, Meygan Martinez, Marissa Piedra, Natalia Reyes, Priscilla Reyes, Pamela Saenz Portillo, Brianna Sanchez, Paola Suarez, Grecia Torres, Denise Borja, Danielle Martinez, Manuel Llano, Germán Rosas-Acosta

During the 2015-2016 academic year, our Phage Hunters group at UTEP aimed to characterize novel bacteriophages infecting a laboratory strain of Arthrobacter sp. Arthrobacteria are typically found in humid environments; therefore, we searched for Arthrobacteriophages in two areas within our region that exhibit substantial differences in humidity: the El Paso, TX, area in the Chihuahuan desert, where forty-two soil samples were harvested, and the Lincoln National Forest in Cloudcroft, NM, where nine samples were collected. Whereas none of the samples collected in El Paso were found to contain phages, one sample collected near Cloudcroft exhibited abundant phage activity. The absence of Arthrobacteriophages in the El Paso area is in marked contrast with the high frequency of Mycobacteriophages reported in this area by our previous Phage Hunters groups.   
Out of the single positive sample from Cloudcroft, upon multiple rounds of plaque purification we obtained nine phage isolates, all of which were analyzed by electron microscopy and DNA restriction fingerprinting. Two morphologically distinct phage types were identified and one phage from each type, namely Piccoletto, a Myoviridae, and Nahla, a Siphoviridae, were selected for DNA sequencing and genomic annotation. According to the sequencing data obtained, Piccoletto and Nahla differed only by a 61 bp sequence that was present in both genomes but that appeared to be duplicated in Nahla. In view of such limited genomic differences between Piccoletto and Nahla, a surprising finding due to their large morphological differences, we considered it essential to further confirm the data. To this end, we first performed a second round of electron microscopy, which produced images consistent with those previously obtained. Second, we explored the genomic region containing the putative 61 bp duplication in Nahla by PCR amplification and DNA sequencing of the PCR products. In these studies, the range of templates used for PCR amplification included the DNAs used for genomic sequencing and the phages used for electron microscopy. These analyses denied the existence of any sequence difference in that region between Piccoletto and Nahla. Finally, we performed restriction analyses of genomic DNAs purified from the phages used for electron microscopy. These analyses revealed an identical DNA restriction profile for both phages.   
Altogether, the data obtained leads us to conclude that the dramatic difference in phage morphology between Piccoletto and Nahla must be attributed to a factor other than genomic sequence differences. We postulate that the morphological differences may be due to differentially regulated phage gene expression involving an epigenetic mechanism. The existence of two morphological stages for the Arthrobacter bacterial host justifies the need for two different morphologies in a phage targeting a single host.