Novel insight into the relationship between genomic sequence and bacteriophage morphology

C. Ontiveros, J. Echeverri, A. Alvidrez, D. Amparan, K. Behrens, R. Bonilla, I. Bustillos, G. Herrera, M. Hidrugo, J. Lujan, M. Martinez, M. Piedra, N. Reyes, P. Reyes, P. Saenz Portillo, B. Sanchez, P. Suarez, G. Torres, D. Borja, D. Martinez, Dr. M. Llano, Dr. G. Rosas-Acosta

ABSTRACT

During the 2015-2016 academic year, our Phage Hunters group at UTEP aimed to characterize novel bacteriophages infecting a laboratory strain of Arthrobacter sp. Arthrobacteriophages are typically found in harsh 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, and the Lincoln National Forest in Cloudcroft, NM. Arthrobacteriophages were isolated from a variety of soil samples located in both areas. To determine if there are differences in the morphologies of bacteriophages that infect the same host, we compared two phage types isolated from soil samples collected in the El Paso area. One phage from each type, namely Piccoletto and Nahla, 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 different in Nahla. In view of such limited genomic differences, we considered it essential to further confirm the data. To this end, we performed two rounds 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 performing a sequence-specific PCR. Two sets of primers were designed within the duplicated region. One primer set used the duplicated sequence as template and the other primer set used the original sequence as template. If the PCR products illustrated in Fig.1 were sequenced by The University of Texas at El Paso, further confirming the synthesis of identical products for both Piccoletto and Nahla template DNA.

MATERIALS AND METHODS

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, and the Lincoln National Forest in Cloudcroft, NM. 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, and the Lincoln National Forest in Cloudcroft, NM. Forty-two soil samples from the El Paso, TX, area were negative for Arthrobacteriophages. One out of nine soil samples from Cloudcroft, NM, yielded two distinct Arthrobacteriophages, Piccoletto and Nahla.

RESULTS

- Sequence data indicated that Piccoletto and Nahla differed from each other by a 61 bp repeat.
- Electron microscopy data showed substantial morphological differences.
- To confirm the data, we analyzed the region containing the 61 bp repeat by PCR and sequencing.

DISCUSSION

The distinct morphologies of phage Piccoletto and phage Nahla, demonstrated that they are morphologically different from each other. Restriction digestion and PCR analysis, shown in Fig. 2, indicated that the two phage types differed from each other by a 61 bp DNA sequence that differed in each genome. Our extensive analysis through restriction digest and PCR strongly indicate that such repeat is not present in Piccoletto, whereas it is present in Nahla. Piccoletto and Nahla differed only by a 61 bp sequence that was present in both genomes but that appeared to be different in Nahla. In view of such limited genomic differences, we considered it essential to further confirm the data. To this end, we performed two rounds 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 performing a sequence-specific PCR. Two sets of primers were designed within the duplicated region. One primer set used the duplicated sequence as template and the other primer set used the original sequence as template. If the PCR products illustrated in Fig.1 were sequenced by The University of Texas at El Paso, further confirming the synthesis of identical products for both Piccoletto and Nahla template DNA.

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