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GENOME ANNOUNCEMENT

Complete Genome Sequence of Temperate Bacteriophage *Aca*ML1 from the Extreme Acidophile *Acidithiobacillus caldus* ATCC 51756

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Development of reproducible genetic tools in the industrially important acidithiobacilli is urgently required. Inducible temperate phages which may be modified *in vitro*, propagated in suitable hosts, and used to transduce relevant genetic information to other strains and/or species are potentially valuable tools in this field of research. In order to address these current limitations, the genome sequence of an inducible temperate *Myoviridae*-like bacteriophage from the *Acidithiobacillus caldus* type strain was annotated and analyzed bioinformatically. Here, we announce the genome sequence of *Aca*ML1 and report major findings from its annotation.

The acidithiobacilli are gammaproteobacteria that are ubiquitous in biomining biotopes, with several characterized species that play key roles in industrial metal recovery (6). To date, no phage capable of infecting members of this group has been identified.

Several members of this group have recently been sequenced (3, 8–10). Bioinformatic analysis of the genome sequence of the moderately thermophilic, sulfur-oxidizing acidophile *Acidithiobacillus caldus* ATCC 51756 predicted the presence of a 59-kb temperate phage located between *srrA* tmRNA and $tRNA^{Met}$. Given the absence of reproducible genetic modification tools for the acidithiobacilli (5), identification and genomic characterization of this novel lysogenic, inducible *Myoviridae* bacteriophage, termed *A. caldus* myovirus-like 1 (*Aca*ML1), potentially capable of infecting and transducing genetic information into members of this genus, represent a major contribution to the field.

Genomic libraries of 4 kb and 40 kb were constructed and sequenced using the random shotgun strategy (7). Assembly of qualified reads, gene modeling, and annotation were performed as previously described (8–10). Curation of the annotation was carried out using conserved protein motif analyses and motif databases, such as Interproscan (http://www.ebi.ac.uk/Tools/pfa/iprscan) and POG (4), and comparative genomic strategies against viral sequence repositories, such as PhAnToMe (http://www.phantome.org).

The *Aca*ML1 genome has 59,353 bp and a GC content of 64.5%, which is higher than the average GC content of its host (61.6%). It is predicted to encode 72 gene products arranged in 10 clusters, 52.8% of which display sequence similarity to known proteins, 34.4% are hypotheticals, and 12.8% present no similarity to any other proteins reported to date. Gene clusters 1 and 2 encode signature proteins implicated in lysogeny establishment and regulation and control of the lysogeny-lytic switch (integrase, excisionase, regulators, primase, RNase endonuclease, methyltransferases). Gene clusters 3 to 9 encode gene products predicted to play a role in viral particle formation and assembly (procapsid shell and maturation protease, baseplate, contractile tail tube, and tail fibers), DNA packaging (terminase, portal protein), and host lysis for viral particle release (holin and endolysin).

The distal gene module in *Aca*ML1 contains three genes encoding an McrBC DNA restriction system and an accompanying modification enzyme (cytosine methylase) and two insertion sequences (IS5, IS21). The presence of RM systems in phages is frequent, playing a relevant role in protection of phage DNA from degradation during phage subversion of the host resources and/or the stabilization of the mobile element within the host genome (e.g., reference 2). These gene cassettes are frequent targets of horizontal gene transfer between bacteria, further facilitated by flanking transposases (e.g., reference 1).

Ongoing studies of the genome of *Aca*ML1 and its life cycle, including its infectivity, latent period, burst size, and host range, will deepen our understanding of its interaction with the *A. caldus* host and provide further insight into its potential as a tool for genetic modification in this group of biotechnologically relevant but genetically recalcitrant microbes.

Nucleotide sequence accession number. The complete genome sequence of *Acidithiobacillus caldus Myoviridae*-like phage *Aca*ML1 is available in GenBank under accession number JX507079.

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