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GENOME SEQUENCES

Draft Genome Sequence of Phocine Herpesvirus 1 Isolated from the Brain of a Harbor Seal

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ABSTRACT Phocine herpesvirus 1 (PhHV-1) is a viral pathogen with high prevalence, morbidity, and mortality in harbor seals. In this study, we used a metagenomic approach to assemble the PhHV-1 genome from the brain tissue of a harbor seal. Here, we present a 119-kb draft genome of PhHV-1 comprising 76 open reading frames.

Phocine herpesvirus 1 (PhHV-1) is a linear double-stranded DNA virus in the subfamily *Alphaherpesvirinae,* genus *Varicellovirus*, that causes high morbidity and mortality in neonatal harbor seals (1, 2). It was discovered in Europe during an outbreak in orphaned young harbor seals, it later emerged in North America in a marine mammal rehabilitation center, and it was responsible for the deaths of almost half of the infected harbor seals (3, 4).

A shotgun metagenomics approach was used to sequence the genome of PhHV-1 from a harbor seal that died from this virus (5). The tissue was extracted from the fresh carcass of a neonatal harbor seal that died on 7 April 2011. Brain tissue was necropsied and frozen by the Marine Mammal Center in Sausalito, CA, in the United States (Table 1). Approximately 0.2 g of tissue was homogenized and extracted with the PowerSoil DNA isolation kit (Mo Bio, CA). DNA quality and quantity were evaluated on an Agilent Bioanalyzer 2100 instrument and a Qubit fluorometer, respectively, at the Center of Genome Research and Biocomputing (CGRB) at Oregon State University (OSU). The sample was then prepared for sequencing with the NexteraXT kit, and the library was sequenced on the Illumina HiSeq 3000 platform using paired-end 150-bp read technology.

Sequenced reads were filtered with fqtrim version 0.9.4 with options -D, -R, -B, -y 10, -q 30, -w, –m, -l 50, and –T, resulting in 427,954,220 reads (6). Reads were then error corrected and normalized with bbnorm-37.85, leaving 144,517,079 paired-end sequences after quality control was performed. Host sequences were then removed by alignment against the Antarctic fur seal (*Arctocephalus gazelle*) genome (doi: 10.5061/dryad.8kn8c.2/1.2) with Bowtie version 2.3.4.3 using the parameters –mp 5, and –local, leaving 21,690,497 paired-end reads (7, 8). Normalized and filtered paired-end reads were used for contig assembly using IDBA_UD version 1.1.3 with –pre_correction, which yielded 945,915 contigs (9). The canine herpesvirus 1 (CHV-1; GenBank accession number KT819633) is the closest relative to PhHV-1; thus, the PhHV-1 contigs were compared to the CHV-1 genome with blastn.2.2.27+ using an expected E value of $10e^{-1}$ (10). The 6 contigs with blast similarities to CHV-1 were then extended with PriceTI v1.2 with parameters 1 1 5, -nc 30, -dbmax 72, -mol 20, -tol 40, -mpi 80, -target 90 2 1 1 (Table 1).

Generally, alphaherpesvirus genomes contain four genomic components, the unique

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TABLE 1 Characteristics of the phocine herpesvirus 1 genome sequencing project with MIGS and MIMS standards[a]

Feature	Description
Taxid[b]	47418
Scientific name	Phocid alphaherpesvirus 1
Common name	Phocid herpesvirus 1
Sample title	Seal5
Sample description	Brain tissue from harbor seal (*Phoca vitulina*)
Investigation type	Metagenome
Project name	Draft genome of phocine herpesvirus 1 infections found in the brain of a harbor seal
Genome sequencing method	Illumina HiSeq 3000
Collection date	7 April 2011
Geographic location (country and/or sea)	California, USA
Geographic location (latitude)	~38.0834°N
Geographic location (longitude)	~122.7633°W
Environment type	Host-associated
Environment (biome)	Seal
Environment (feature)	Brain
Environment (material)	Tissue
Genome assembly	IDBA_UD
Genome coverage (×)	41
Finishing strategy	PriceTI
ENA accession number	ERS1903621

[a]MIGS, minimum information about a genome sequence; MIMS, minimum information about a metagenomic sequence/sample.
[b]Taxid, NCBI taxonomy identification number.

long (U_L), unique short (U_S), terminal and internal inverted repeats long (TR_L/IR_L), and terminal and internal inverted repeats short (TR_S/IR_S). For alphaherpesvirus, these genetic regions have a generic genome layout as follows: TR_L-U_L-IR_L-IR_S-U_S-TR_S. Our analysis resulted in 3 contigs with lengths of 98.2 kb (TR_L-U_L-IR_L), 15.7 kb (IR_S[partial]-U_S-TR_S[partial]) and 5.3 kb (IR_S[partial]-TR_S[partial]) that totaled 119,153 kb, with an estimated genome size of ~124.4 kb. The average GC content of the 3 contigs was 34.4%. Annotation of 76 open reading frames (ORFs) identified by Prokka.v1.12 resulted in a total of 72 predicted protein-encoding genes (11), with 60 gene annotations found in the U_L and 8 in the U_S and 4 diploid genes (a total of 8 ORFs) in the TR_S/IR_S.

Data availability. The draft genome sequence reported here was deposited in the European Nucleotide Archive (ENA) under the accession number ERS1903621 and the sequence reads were deposited under accession number ERX3182993. The deposited sequences were quality filtered, and the adapters were removed.

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