



Complete Genome Sequences of the Index Isolates of Two Genotypes of Pacific Salmon Paramyxovirus

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ABSTRACT We report here the genome sequences of two index strains of Pacific salmon paramyxovirus isolated in 1982 and 1983 from adult salmon in Oregon. The isolates are most closely related to Atlantic salmon paramyxovirus, the type species of the genus *Aquaparamyxovirus*, but are sufficiently distinct to be considered two genotypes of a novel species.

In 1982 and 1983, routine testing of Chinook salmon (*Oncorhynchus tshawytscha*) returning to hatcheries on the coast of Oregon produced two isolates of a novel virus (1). The isolates had characteristics of members of the *Paramyxoviridae*, including the ability to hemagglutinate erythrocytes and initiate persistent infections in cell lines, but they appeared to be avirulent in four species of Pacific salmon (2). Subsequently, isolates of this Pacific salmon paramyxovirus (PSPV) were recovered from many stocks of adult salmon along the Pacific coast of North America, and sequence comparisons of a region of the viral polymerase showed the presence of two genetic sublineages that appeared to be cocirculating in the North Pacific Ocean (3). The analysis also showed that the virus was most closely related to the Atlantic salmon paramyxovirus (ASPV) obtained from salmon with proliferative gill disease (4–6). Currently, ASPV is the type species of the genus *Aquaparamyxovirus* and was given the formal species name *Salmon aquaparamyxovirus* (https://talk.ictvonline.org/taxonomy).

To determine the genome sequence of the index isolate representing each PSPV sublineage, we performed Sanger and Illumina sequencing on RNA extracted from the 1983 Trask River isolate (sublineage A) and the 1982 Yaquina River isolate (sublineage B) propagated in the CHSE-214 cell line, as previously described (2). Genomic RNA and library preparation for Illumina sequencing (MiSeq version 2; 2 imes 150 bp) were performed as described previously (7). Briefly, RNA was extracted (QIAamp viral RNA minikit; Qiagen) from filtered (0.8-µm pore size) and nuclease-treated cell culture supernatant, followed by whole-transcriptome amplification (WTA2; Sigma-Aldrich) and library preparation (Nextera XT preparation kit; Illumina), following the manufacturers' instructions. We mapped adaptor-trimmed Illumina reads (CLC Genomics Workbench version 11) for the Trask River isolate (1,529,148 raw reads) using a reference genome composed of the full ASPV genome and extant partial sequences of the Trask River isolate from GenBank (accession numbers EU156171, AY536862, and JF795583). Sanger sequencing was used to fill in gaps or clarify ambiguities, and rapid amplification of cDNA ends (RACE) was used to determine the 3' and 5' terminal sequences, as described previously (8). The complete Trask River genome was used to map Illumina reads of the Yaquina River isolate (1,686,622 raw reads), with gaps and termini determined as described above. In total, 1,337,886 (87%) and 1,196,563 (71%) reads mapped to the finished Trask and Yaquina genomes, respectively.

The genomes of the Yaquina 1982 and Trask 1983 isolates of PSPV were both 16,302 nucleotides in length, obeying the "rule of six" in which paramyxovirus genomes are evenly divisible by six. Sequence alignments of the full-length genomes showed that

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Received 6 November 2018 Accepted 8 February 2019 Published 7 March 2019 the two strains shared an 87.5% overall nucleotide identity with no gaps, but they were only 60.0% identical with ASPV (with 151 to 165 gaps in the alignments). The genome organization for both isolates was the same as for ASPV, with six genes in the order 3'-N-P-M-F-HN-L-5'. Within the P gene, an alternate open reading frame (ORF) encodes the putative C protein. The seven ORFs of the two strains had amino acid identities that ranged from 95.9% (P) to 99.3% (L), but these were only 33.2% (P) to 74.4% (L) identical to those of ASPV. Thus, the isolates appear to represent separate genotypes of a new virus species in the genus *Aquaparamyxovirus*.

Data availability. The complete genome sequences (and raw sequence reads) of the Trask River 1983 and Yaquina River 1982 isolates of Pacific salmon paramyxovirus have been deposited in GenBank under accession numbers MH900516 (SRA accession number SRX5329359) and MH900517 (SRA accession number SRX5329360), respectively.

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