





Genome Sequences of Vesicular Stomatitis Indiana Viruses from the 2019 Outbreak in the Southwest United States

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ABSTRACT We report the genomes of three vesicular stomatitis Indiana virus (VSIV) isolates collected from naturally infected bovines in Wyoming and Colorado during the 2019 outbreak in the United States. These genomes support molecular diagnostic efforts and provide data on the spread and ecology of VSIV in the United States.

esicular stomatitis (VS) is the most common vesicular disease of livestock in the Americas, infecting horses, pigs, and cattle. In pigs and cattle, the clinical signs are similar to the highly contagious foot-and-mouth disease (FMD) (1). VS is caused by the arthropod-borne VS virus (VSV), a commonly studied virus from the Vesiculovirus genus of the Rhabdoviridae family. The ~11-kb negative-sense, single-stranded RNA genome of VSV encodes five viral structural proteins, the nucleoprotein (N), phosphoprotein (P), matrix protein (M), and glycoprotein (G) and the large RNA-dependent RNA polymerase

Vesicular stomatitis New Jersey virus (VSNJV) and VSIV are the two principle serotypes of VSV, with VSNJV causing the vast majority of clinical cases in livestock. VSV is not endemic in the United States but is endemic from the southern states of Mexico through Central America and into northern South America (4). Prior to 2019, the last occurrence of VSIV in the United States was in 1998 (5). The 2019 VSIV index case was identified in Texas in June, and by the end of 2019, cases were confirmed in seven additional states (New Mexico, Colorado, Wyoming, Oklahoma, Nebraska, Utah, and Kansas) (https://www.aphis.usda.gov/aphis/ourfocus/animalhealth/animal -disease-information/cattle-disease-information/vsv-reports).

Field samples (Table 1) from cattle displaying clinical signs indicative of VS were submitted to the Foreign Animal Disease Diagnostic Laboratory at the Plum Island Animal Disease Center in New York and sequenced as previously described (6). Briefly, viral isolates were recovered from swabs or epithelial lesion tissues after one passage in Vero cells. Total nucleic acid was extracted from the supernatant using the MagMax Pathogen RNA/DNA extraction kit (Applied Biosystems) on a MagMAX Express platform (Applied Biosystems). First-strand synthesis was performed using SuperScript III reverse transcriptase (Invitrogen) with random primers. Second-strand synthesis was performed using Sequenase enzyme (Affymetrix) and then amplified with Tag polymerase (Clontech). Sequencing libraries were constructed using the Nextera XT kit (Illumina) and sequenced with a 500-cycle MiSeq sequencing kit v2 on the MiSeq System (Illumina). A custom pipeline (https://github.com/rwbarrette/RWB_NGS_scripts/blob/ master/Multi_Reference_Guided_Assembly.py) written in Python v 2.7.8 was used to produce the consensus sequences from quality-trimmed reads, and the resulting fastq files were mapped to a VSIV genome as the reference scaffold (GenBank accession number NC_001560). The pipeline used the Burrows-Wheeler Aligner (BWA) v 0.7.12 for

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TABLE 1 Sampling locations, dates, sequencing metrics, and accession numbers for the sequences in this report

	Location	Date of collection	Infected	Total no.	Avg	GC	GenBank	SRA	Genome
Isolate	(city, state)	(day-mo-yr)	species	of reads	coverage (\times)	content (%)	accession no.	accession no.	length (nt)
IN0919WYB1	Wheatland, WY	5-Sep-19	Bovine	1,708,558	7,490.79	41.80	MT437284	SAMN14931266	11,161
IN0919WYB2	Garland, WY	9-Sep-19	Bovine	475,405	5,777.96	41.80	MT437283	SAMN14931267	11,161
IN0919COB	Lyons, CO	18-Sep-19	Bovine	1,893,332	7,600.35	41.80	MT437285	SAMN14931268	11,161

initial reference-guided assembly using default parameters. SAMtools and BCFtools v 1.1 were used for processing, while the mpileup tool was used to perform the consensus assembly.

The three isolates displayed the same genomic length (11,161 nucleotides [nt]), organization, and GC content (Table 1). The sequences share >99.9% identity with each other. The closest BLASTn match for all three sequences was a genome from the 1998 U.S. outbreak (GenBank accession number NC_038236.1) with 98.88% to 98.90% sequence identity.

Since VS presents similarly to FMD in cattle and pigs, a rapid differential diagnosis is extremely important. These sequences from the first VSIV outbreak in the United States in over 20 years should prove extremely valuable in evaluating diagnostic assays. Additionally, they might provide insights into viral introduction and transmission and may help inform mitigation strategies.

Data availability. The consensus sequences have been deposited in GenBank under accession numbers MT437283 to MT437285. The raw sequencing reads are available in the NCBI Sequence Read Archive under accession number PRJNA633040.

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