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GENOME ANNOUNCEMENT

Complete Genome Sequence of Porcine Reproductive and Respiratory Syndrome Virus Isolated from Piglet Stool Samples

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WUH4 is a highly pathogenic North American porcine reproductive and respiratory syndrome virus (PRRSV). Unlike previous PRRSV isolates, which were mainly recovered from sera or tissues, WUH4 was isolated from a piglet stool sample. Here we announce its complete genome sequence.

orcine reproductive and respiratory syndrome (PRRS) is an emerging viral infectious disease characterized by severe reproductive failure in sows and respiratory distress in piglets and growing pigs (6, 9). The causative agent, PRRS virus (PRRSV), is a single-stranded positive-sense RNA virus classified within the family Arteriviridae (3). Since its emergence in the late 1980s, PRRS has continuously been a threat to the global swine industry, causing high economic losses (6, 11, 15). Because PRRSV has a strong tropism for monocyte/macrophage lineages in vivo (4, 12) and PRRSV infection results in persistent viremia (1), previous PRRSV isolates were mainly recovered from serum, lung, or brain (2, 7, 8, 13, 14). PRRSV strain WUH4 was accidentally isolated from a piglet stool sample in China in 2011. An obvious cytopathic effect (CPE), characterized by clumping, shrinkage, and detachment, could be observed in WUH4-infected porcine alveolar macrophages (PAMs). To understand the diversity and the evolutionary characteristics of WUH4, we determined its complete genome sequence.

Fourteen pairs of oligonucleotide primers were designed based on the sequences of PRRSV strains VR-2332 (2) and JXA1 (13) to amplify the different regions of WUH4 genome. The PCR products were cloned into pMD18-T vector (TaKaRa) and sequenced with an ABI3730XL genome sequencer. The terminal sequences were acquired by using a kit for rapid amplification of cDNA ends (RACE) (Clontech, Japan). The sequence data of strain WUH4 were assembled into one contiguous sequence of 15,339 nucleotides, including the poly(A) tail. The genome of WUH4 shared 89.6% and 99.7% sequence identity with the representative North American strains of PRRSV and only 58.1% sequence identity with the European prototype Lelystad virus (LV) (10), indicating that WUH4 belongs to the North American genotype of PRRSV. Phylogenetic analyses based on the genome sequence also supported this conclusion. Notably, the nucleotide sequence and genome organization of WUH4, especially a discontinuous deletion of 30 amino acids (aa) (at aa 481 and 533 to 561) in nonstructural protein 2 (Nsp2)-coding regions, were highly similar to those of strain JXA1, the representative isolate of the highly pathogenic PRRSV that emerged in China in 2006 (13, 14). Furthermore, the 5' untranslated region (UTR) and 3' UTR and the predicted nonstructural proteins encoded by open reading frames (ORFs) 1a and 1b of WUH4 showed more than 99.5% identity with JXA1. The predicted structural proteins GP2, M, and N of WUH4 showed 100% amino acid identity with JXA1. However, compared with JXA1 and the attenuated vaccine strain (5), mutation at S³⁷

(S37P) in the primary neutralizing epitope of GP5 was found in WUH4, indicating that WUH4 may possess the potential to circumvent immune responses induced by currently used vaccines.

The availability of the genome sequence of WUH4 will facilitate future investigations of the transmission of PRRSV via the fecal-oral route. Furthermore, PRRSV can be recovered from stool samples, which raises the issue of how well PRRSV replicates in the gastrointestinal tract.

Nucleotide sequence accession number. The genome sequence of PRRSV strain WUH4 has been deposited in GenBank under accession number JQ326271.

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