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# Complete Genome Sequence of an Overattenuated Highly Pathogenic Porcine Reproductive and Respiratory Syndrome Virus

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GENOME ANNOUNCEMENT

# Complete Genome Sequence of an Overattenuated Highly Pathogenic Porcine Reproductive and Respiratory Syndrome Virus

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**JXA1-P170 is an overattenuated highly pathogenic porcine reproductive and respiratory syndrome virus (HP-PRRSV) that has been passaged *in vitro* 170 times. Vaccination with JXA1-P170 cannot protect pigs against JXA1 challenge. Compared with the parental virus JXA1, JXA1-P170 contains 1 nucleotide (nt) deletion and 113 nt mutations leading to 59 amino acid substitutions. Here we announce the first complete genome sequence of the overattenuated HP-PRRSV.**

Porcine reproductive and respiratory syndrome virus (PRRSV) is an enveloped, positive-sense, single-stranded RNA virus belonging to the *Arterivirus* genus, family *Arteriviridae*, order *Nidovirales* (2, 10). Highly pathogenic PRRSV (HP-PRRSV) is a mutant of PRRSV containing a novel discontinuous 30-amino-acid (30-aa) deletion in nonstructural protein 2 (Nsp2) (7, 12). HP-PRRSV is the etiological agent of HP-PRRS epidemics, which first emerged in southern China in 2006; the virus is currently widespread in China and several Southeast Asian countries, causing enormous economic losses (1, 3, 6, 9, 12). Live attenuated PRRSV vaccines based on serial *in vitro* passage are efficient tools that can be used to protect pigs against PRRSV infection (8, 13). However, little is known about the genomic characteristics of the overattenuated PRRSV. Here we report the complete genome sequence of JXA1-P170, which is an overattenuated HP-PRRSV.

JXA1-P170 is a product that resulted from 170 continuous passages of strain JXA1 (GenBank accession no. EF112445) in Marc-145 cells, using the same assay as previously reported (5). Eighteen pairs of primers were used to amplify 18 overlapped fragments of the JXA1-P170 genome (5). PCR products were cloned into pGEM-T Easy vector (Promega), sequenced three times with an ABI Prism 3730 sequencer (Applied Biosystems), and assembled into the full-length sequence with SeqMan software (DNASTAR Inc.). The genome of JXA1-P170 is 15,319 bp, excluding the 3′ poly(A) tail, and shares 99.26% nucleotide identity with the genome of the JXA1 parental virus. In addition to the 30-aa deletion in Nsp2, JXA1-P170 has 1 nucleotide (nt) deletion at position 115 within the 5′ untranslated region (5′UTR). In addition, it has two substitutions ($T_{116}C$ and $A_{124}G$) in the 3′UTR. Compared with JXA1, JXA1-P170 has 113 nt mutations that result in 59 aa substitutions. Compared with JXA1-P80 (GenBank accession no. FJ548853), JXA1-P170 has 30 nt mutations leading to 22 aa replacements.

Vaccination with JXA1-P80 can provide sufficient protection against JXA1 infection; however, when pigs were inoculated with JXA1-P170 for 28 days and then challenged with JXA1, they still showed high fever and clinical signs similar to those seen in pigs without vaccination. These results indicate that JXA1-P170 is overattenuated and cannot provide effective protection in pigs. It is generally believed that the 5′UTR of PRRSV plays vital roles in viral replication, mRNA transcription, and protein translation (4); therefore, the guanine deletion in the 5′UTR of JXA1-P170 might affect its infectivity in the pig. The substitutions in the 3′UTR might also affect viral replication (11). The mutation differences between JXA1-P170 and JXA1 could help researchers to explore the virulence change, and the mutation differences between JXA1-P170 and JXA1-P80 should be crucial for understanding their differences in inducing host immune responses.

This is the first report of an overattenuated HP-PRRSV strain. The complete genome of JXA1-P170 should facilitate future research aimed at elucidation of the mechanisms of pathogenicity and immunogenicity changes from JXA1 to JXA1-P80 and then to JXA1-P170 and should also contribute to the production of a live-attenuated PRRSV vaccine with higher efficacy.

**Nucleotide sequence accession number.** The complete genome sequence of JXA1-P170 has been deposited in GenBank under accession number JQ804986.

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