

Complete Genome Sequence of Porcine Reproductive and Respiratory Syndrome Virus Strain QY2010 Reveals a Novel Subgroup Emerging in China

Yuxiu Deng,^b Yongfei Pan,^a Dongdong Wang,^a Qingfeng Zhou,^a Yingzuo Bi,^b Feng Chen,^b and Yanhua Song^a

Guangdong Wen's Group Academy, Guangdong Wen's Foodstuffs Group Co., Ltd., Xinxing, Guangdong, China,^a and College of Animal Science, South China Agricultural University, Guangzhou, China^b

QY2010 is a highly pathogenic North American-type porcine reproductive and respiratory syndrome virus (PRRSV). The complete genome sequence shows that QY2010 shares low sequence identity (60 to 88.7%) to all known PRRSV isolates. Phylogenetic analyses further reveal that QY2010 constitutes a novel subgroup within the North American genotype of PRRSV.

Porcine reproductive and respiratory syndrome (PRRS) is the cause of an economically important swine disease that has been devastating the swine industry since the late 1980s (4, 8, 13). The etiologic agent, PRRS virus (PRRSV), is a positive-strand RNA virus belonging to the family *Arteriviridae* (2). PRRSVs are genetically and antigenically highly heterogeneous and are divided into European and American genotypes (10, 12). Remarkable sequence differences have been found among isolates of the same genotype, particularly in the Nsp2 and open reading frame 5 (ORF5) genes (3, 9). PRRSV was first reported in 1996 in China. Most Chinese isolates belong to the American genotype and can be further classified into four subgroups based on the whole genome (1, 5). Under the immune pressure caused by the current control strategy, in which attenuated vaccines are widely used, PRRSVs are geared for rapid variation through mutation or recombination (6, 7, 11, 14), resulting in new isolates with different levels of pathogenicity and virulence.

Here, we report a PRRSV strain, QY2010, isolated from lung samples of a piglet with respiratory symptoms and PRRSV positive by reverse transcription (RT)-PCR, in South China in 2010. Experimental animal infection revealed that all piglets infected with QY2010 developed typical clinical signs of PRRS, including labored breathing, lameness, emaciation, and depression, and 3 of 10 piglets had died at the end of the experiment, confirming that QY2010 was a highly pathogenic PRRSV.

Fourteen pairs of oligonucleotide primers to amplify the different regions of the QY2010 genome were designed based on the sequences of PRRSV strains VR-2332 and JXA1. The PCR products were purified, cloned into the pMD18-T vector (TaKaRa, Japan), and sequenced with an automated genome sequencer (Genetic Analyzer 3730XL; Applied Biosystems). The terminal sequences were acquired by using a kit for rapid amplification of cDNA ends (RACE) (Clontech, Japan). Sequence alignment was performed using Clustal X 2.1 (16). A phylogenetic tree was constructed using MEGA 4 (15).

The complete genome of the QY2010 strain is 15,514 nucleotides in length, excluding the poly(A) tail. Its genome organization is similar to those of other reported PRRSV genomes. Interestingly, a continuous insertion of 108 nucleotides was found in the Nsp2 gene (at positions 3771 to 3878), and the insertion shows no homology with any known proteins. Its origin needs to be identified. The genome of QY2010 exhibited 86.3% to 88.7% nucleotide identity to all North American strains of PRRSV and only 60.0%

nucleotide identity with the European prototype Lelystad virus (LV). Further phylogenetic analyses based on the genome indicated that QY2010 constitutes a new subgroup, an addition to the previous 4 to 5 subgroups within the North American subtypes. These findings reveal the emergence of a new subgroup of American subtypes in China, and the question of how QY2010 acquired such a large mutation within its genome needs to be investigated in the future. The genome data for QY2010 will facilitate future investigations of the evolutionary characteristics and molecular pathogenesis of PRRSV.

Nucleotide sequence accession number. The genome sequence of PRRSV strain QY2010 has been deposited in GenBank under accession number [JQ743666](https://www.ncbi.nlm.nih.gov/nuclseq/JQ743666).

ACKNOWLEDGMENT

This work was supported by a Provincial Key Science and Technology Projects grant (2008A 020100020) from the Science and Technology Department of Guangdong Province, China.

REFERENCES

1. An TQ, et al. 2010. Origin of highly pathogenic porcine reproductive and respiratory syndrome virus, China. *Emerg. Infect. Dis.* 16:365–367.
2. Dea S, Gagnon CA, Mardassi H, Pirezadeh B, Rogan D. 2000. Current knowledge on the structural proteins of porcine reproductive and respiratory syndrome (PRRS) virus: comparison of the North American and European isolates. *Arch. Virol.* 145:659–688.
3. Forsberg R, et al. 2002. The genetic diversity of European type PRRSV is similar to that of the North American type but is geographically skewed within Europe. *Virology* 299:38–47.
4. Keffaber KK. 1989. Reproductive failure of unknown etiology. *Am. Assoc. Swine Pract. Newslett.* 1:1–10.
5. Li B, et al. 2010. The genomic diversity of Chinese porcine reproductive and respiratory syndrome virus isolates from 1996 to 2009. *Vet. Microbiol.* 146:226–237.
6. Li B, et al. 2009. Recombination in vaccine and circulating strains of porcine reproductive and respiratory syndrome viruses. *Emerg. Infect. Dis.* 15:2032–2035.
7. Liu D, et al. 2011. Recombination analyses between two strains of porcine reproductive and respiratory syndrome virus in vivo. *Virus Res.* 155:473–486.

Received 20 April 2012 Accepted 23 April 2012

Address correspondence to Yanhua Song, yanhua_song@163.com.

Y.D. and Y.P. contributed equally to this work.

Copyright © 2012, American Society for Microbiology. All Rights Reserved.

doi:10.1128/JVI.00977-12

8. Lunney JK, Benfield DA, Rowland RR. 2010. Porcine reproductive and respiratory syndrome virus: an update on an emerging and re-emerging viral disease of swine. *Virus Res.* 154:1–6.
9. Mateu E, Martin M, Vidal D. 2003. Genetic diversity and phylogenetic analysis of glycoprotein 5 of European-type porcine reproductive and respiratory virus strains in Spain. *J. Gen. Virol.* 84:529–534.
10. Meng XJ. 2000. Heterogeneity of porcine reproductive and respiratory syndrome virus: implications for current vaccine efficacy and future vaccine development. *Vet. Microbiol.* 74:309–329.
11. Murtaugh MP, Yuan S, Faaberg KS. 2001. Appearance of novel PRRSV isolates by recombination in the natural environment. *Adv. Exp. Med. Biol.* 494:31–36.
12. Nelsen CJ, Murtaugh MP, Faaberg KS. 1999. Porcine reproductive and respiratory syndrome virus comparison: divergent evolution on two continents. *J. Virol.* 73:270–280.
13. Neumann EJ, et al. 2005. Assessment of the economic impact of porcine reproductive and respiratory syndrome on swine production in the United States. *J. Am. Vet. Med. Assoc.* 227:385–392.
14. Song J, Shen D, Cui J, Zhao B. 2010. Accelerated evolution of PRRSV during recent outbreaks in China. *Virus Genes* 41:241–245.
15. Tamura K, Dudley J, Nei M, Kumar S. 2007. MEGA4: Molecular Evolutionary Genetics Analysis (MEGA) software version 4.0. *Mol. Biol. Evol.* 24:1596–1599.
16. Thompson JD, Gibson TJ, Plewniak F, Jeanmougin F, Higgins DG. 1997. The CLUSTAL_X windows interface: flexible strategies for multiple sequence alignment aided by quality analysis tools. *Nucleic Acids Res.* 25:4876–4882.