

Complete Genome Sequence of a Recombinant Nephropathogenic Infectious Bronchitis Virus Strain in China

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Recently, nephropathogenic infectious bronchitis virus (IBV) outbreaks have occurred in commercial broiler flocks and have been associated with a high incidence and morbidity in China. The CK/CH/Zhejiang/06/10 strain (IBV-YX10) was isolated from a 12-day-old broiler chicken in a flock of chickens with swollen speckled kidneys and distended ureters filled with uric acid in China in 2010. Here we reported the complete genomic sequence of the IBV-YX10 which was a natural recombinant nephropathogenic infectious bronchitis virus strain. These findings will contribute additional insights into the molecular characteristics of evolving IBV genomes and the need for effective control of IBV in China.

Infectious bronchitis (IB) is a serious and highly contagious disease of chickens, caused by infectious bronchitis virus (IBV), which is enveloped and belongs to the type species of the *Coronaviridae* family (10). Recently, the outbreaks of IBV infection have been persistent but infrequent in vaccinated broiler flocks and have been associated with a high incidence and morbidity in China (2, 3, 5, 7, 9, 14). We conducted a viral surveillance program from 2004 to 2011 and isolated more than 300 field IBVs from broilers or broiler breeder flocks vaccinated with a classical IBV vaccine in China (flocks in different provinces), and the results of the epidemiological analysis suggested that nephropathogenic IBVs had become more and more prevalent (2, 3, 7, 13). Natural genetic recombination between strains classified into different genetic groups has occurred and may have caused the emergence of new IBV strains (1, 4).

The CK/CH/Zhejiang/06/10 (IBV-YX10) strain was isolated from a 12-day-old broiler chicken in a flock of chickens with swollen speckled kidneys and distended ureters filled with uric acid in Zhejiang province of China in 2010. The IBV-YX10 strain was a highly virulent virus strain that caused high morbidity in the clinic and was used as the vaccine strain against nephropathogenic IBV infection in China. The whole genome of IBV-YX10 was amplified by reverse transcription-PCR (RT-PCR), cloned into the pMD19-T vector (TaKaRa), and sequenced three times. Sequence alignment was performed with Clustal X (BioEdit version 7). Phylogenetic trees were constructed using the MEGA 5 program (11). Recombination analysis was carried out by Simplot version 3.5.1 (6) and the recombination detection program RDP4 v.4.1.3 (8).

The complete genome of IBV-YX10 consisted of 27,674 nucleotides (nt), and the genome consisted of six genes (12). The order of the six genes was as follows: 5' untranslated region (5'UTR)-1a/ab-spike-3a-3b-envelope (3c)-membrane-4b-4c-5a-5b-nucleocapsid-3'UTR (12). The spike (S) protein gene of IBV-YX10 was 3,471 nucleotides long (1,156 amino acids [aa]), which was a glycoprotein on the virus envelope surface and a determinant of the IBV serotype. The cleavage site on the spike protein of IBV-YX10 was HRRRRR, which represented the majority of the IBVs in China (2, 3, 15). Phylogenetic analysis of the S1 gene indicated that IBV-YX10 belonged to genotype I, designated A2-like group or QXIBV-type viruses (4), which had been circulating in China. The whole-genome sequence of IBV-YX10 showed 86.2.0% to 96.4%

homology with the IBV sequences in GenBank but only 86.2%, 86.3%, and 86.7% homology with vaccine strains H52, H120, and M41 (vaccine strains used thus far in China), respectively. Recombination analysis indicated that IBV-YX10 was a potential recombinant strain between the LX4 and GX-YL9 strains, and its major parent was the GX-YL9 strain. The S1 gene, 3a, 3b, and 3c, M, 4b, and 4c, and 5a gene were derived from LX4, which was isolated from Xinjiang province in China in 1999 and belonged to genotype I, and the other genes were derived from GX-YL9, which was isolated from Guangxi province of China in 2007 (9) and belonged to genotype VI. The results indicated that the field IBV-YX10 strain in China had undergone genetic recombination.

Nucleotide sequence accession number. The complete genome sequence of the IBV-YX10 isolate was submitted to GenBank and assigned accession number [JX840411](https://doi.org/10.1128/JVI.02575-12).

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