Complete Genome Sequence of an H3N2 Canine Influenza Virus from Dogs in Jiangsu, China

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A canine influenza virus (CIV) strain of avian origin designated A/Canine/Jiangsu/06/2010 (H3N2) was isolated from dogs exhibiting severe respiratory disease in Jiangsu, China. We announce the complete genome sequence of this viral strain and report major findings from the genomic analysis. This sequence will help us understand the molecular characteristics and evolutionary of H3N2 CIV in China.

Canine influenza virus (CIV) is a newly identified, highly contagious respiratory pathogen of dogs. In 2004, an equine-origin H3N8 influenza virus was first isolated from racing greyhounds affected with respiratory disease in Florida (1). Subsequent outbreaks were reported, and the infection rapidly disseminated across the United States. In 2007, a different subtype of influenza virus, H3N2, caused an outbreak of canine respiratory disease in South Korea (7). This virus appears to be entirely of avian origin, but can be transmitted between dogs. This was the first time that a low-pathogenicity avian influenza virus (AIV) was found to cause respiratory disease in dogs. The emerging pathogen causes considerable concerns for both veterinary and public health.

In this study, an H3N2 strain, named A/Canine/Jiangsu/06/2010, was isolated from dogs in Jiangsu Province, China. We analyzed the complete genome sequence of the viral strain and investigated its molecular characteristics. Viral RNAs were extracted by the use of TRIzol reagent from infectious allantoic fluid from embryonated chicken eggs (3). Standard reverse transcription-PCR was performed with primers specific for influenza virus (2). The PCR products were cloned into the pMD19-T vector (TaKaRa, Dalian, China) and sequenced by the Invitrogen Company, Shanghai, China.

The complete genome of the strain consists of eight segments of negative-sense single-stranded RNA molecules, including PB2, PB1, PA, HA, NP, NA, M, and NS. The full lengths of the segments are 2,341, 2,341, 2,233, 1,765, 1,565, 1,473, 1,027, and 890 nucleotides, respectively. The eight genes encode proteins with the following amino acid lengths: PB2, 759; PB1, 757; PA, 716; HA, 566; NP, 498; NA, 471; M1, 252; M2, 97; NS1, 230; and NS2, 121. All eight genes exhibit the highest homology (>99%) to the canine-derived subtype H3N2 influenza virus isolated in cats from South Korea (6). The strain has Q226 and G228 at the receptor binding site in HA protein, which suggests that it is an avian-origin influenza virus. The amino acid sequence at the cleavage site in the HA molecule is PERQTR ↓ G, with the characteristic of low-pathogenicity AIV. Analysis of potential glycosylation sites of the isolate reveals that there are 7 potential N-linked glycosylation sites in HA (positions 22, 38, 45, 81, 165, 285, and 483), while there are 6 in NA (positions 69, 88, 148, 202, 236, and 404). Significantly, a two-amino-acid insertion is found at the end of the NA stalk. Interestingly, artificial inoculation of BALB/c mice showed the

CIV isolate has a broad tissue tropism and causes lesions in pulmonary and extrapulmonary organs (5). Similar results were found in beagle models (data not shown). These observations are distinct from only respiratory tract disease without systemic infection observed in dogs experimentally infected with South Korean H3N2 CIV (4, 7, 8).

The genome data are useful for analyses of evolutionary characteristics of H3N2 CIV and valuable for understanding the mutation frequencies associated with replication of the virus in the canine host.

Nucleotide sequence accession numbers. The genome sequences of A/Canine/Jiangsu/06/2010 (H3N2) have been deposited in GenBank under accession no. [JN247616](#) to [JN247623](#).

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