

Complete Genome Sequence Analysis of Human Echovirus Type 30 Isolated in China

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We report here the complete genome sequence of a human echovirus type 30 strain ECV30/GX10/05 isolated in Guangxi, China, in 2010. Phylogenetic analysis showed that ECV30/GX10/05 was closely related to a Korean strain isolated in 2008. The sequence information will help in an understanding of the molecular epidemiology and evolution of echovirus.

Human echovirus type 30 (EC30) is a member of the genus *Enterovirus* in the family *Picornaviridae* and belongs to human enterovirus subgroup B (HEV-B) together with 60 other serotypes (7). EC30 is one of the most commonly isolated enterovirus serotypes in multiple meningitis outbreaks (2). In recent decades, EC30-associated meningitis epidemics have been documented throughout the world: in China (17, 18), the United States (6, 10), and many European countries (12–15). An unusual outbreak of EC30-associated aseptic meningitis occurred in Jiangsu Province in China in 2003, which caused increased attention by public health entities (17). EC30 was also found to be involved in several outbreaks of aseptic meningitis and hand, foot, and mouth disease in Zhejiang, Shandong, and Guangxi provinces (3, 4, 16). However, only two EC30 strains from mainland China were recorded in GenBank 10 years ago (3, 17).

In the present study, we report the complete genome sequence of an EC30 strain, ECV30/GX10/05, isolated from fecal samples in Guangxi Province, China, in 2010 (4). Complete genome sequencing was performed according to our previously adopted strategies for coxsackievirus B5 (5). Briefly, viral RNA was extracted from viral culture with an RNeasy minikit (Qiagen), and cDNA was produced by using Moloney murine leukemia virus (M-MLV) reverse transcriptase (TaKaRa) with an oligo(dT) primer. Seven primer pairs were used to generate the amplicons spanning the entire viral genome. The 5' and 3' terminals of the genome were determined using a rapid amplification of cDNA ends (RACE) kit (Roche). All sequencing was carried out using an ABI 3730 Sanger-based genetic analyzer, and sequences were assembled using DNASTAR Lasergene 7.0. Sequence alignment was performed using ClustalX 2.1 and MegAlign. Phylogenetic trees were constructed using MEGA 5.0 software.

The complete genome of EC30 strain ECV30/GX10/05 consists of 7,432 nucleotides and encodes a single open reading frame of 6,585 bases coding for structural and nonstructural proteins flanked by untranslated regions (UTR) at the 5' and 3' ends. The 5' UTR contains 744 bp, and the 3' UTR contains 103 bp. Phylogenetic analysis based on the VP1 or complete genome sequence was conducted using the neighbor-joining method. The results showed that ECV30/GX10/05 was clustered with EC30 strain Kor08-ECV30 (JN704615), isolated in 2008 (1). ECV30/GX10/05 belongs to a distinct genetic lineage with Kor08-ECV30 based on the phylogenetic trees (4). The nucleotide and amino acid homologies of ECV30/GX10/05 with Kor08-ECV30 were 96.6%. The nucleotide homologies with two other Chinese isolates, Echo30/

Zhejiang/17/03/CSF (DQ246620) and FDJS03_84 (AY948442), were 83.5 and 83.3%, and the amino acid homologies were 82.9% and 82.7%, respectively. Preliminary bootscan and similarity analysis using SimPlot 3.5.1 indicated that the EC30 Guangxi strain may have originated from a recombination of Chinese EC30 strains reported in 2003 with other group B human enteroviruses at nonstructural regions (8, 9, 11).

The complete genome sequence of ECV30/GX10/05 presented here will facilitate future investigations of the molecular characteristics of EC30 and help elucidate its recombination and phylogenetic relationship to other enteroviruses.

Nucleotide sequence accession number. The complete genomic sequence of EC30 Guangxi strain ECV30/GX10/05 was deposited in the GenBank database under accession no. [JX854435](https://www.ncbi.nlm.nih.gov/nuccore/JX854435).

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