



## Article Isolation, Identification, and Genetic Phylogenetic Analysis of Two Different Genotypes of Bovine Parainfluenza 3 Virus in China

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**Abstract:** Bovine parainfluenza virus 3 (BPIV3) is one of several viruses that contribute to bovine respiratory disease complex (BRDC). During this study, isolation of BPIV3 was attempted from 20 PCR-positive swabs by Madin-Darby Bovine Kidney (MDBK) cells. Nine samples showed obvious cytopathic lesions identified as BPIV3 by reverse-transcription polymerase chain reaction amplification and sequencing. The genomes of isolates XJ21032-1 and XJ20055-3 were sequenced using Illumina sequencing technology and determined to have lengths of 15,512 bp and 15,479 bp, respectively. Phylogenetic analysis revealed that isolate XJ21032-1 was genotype B, and isolate XJ20055-3 was genotype C. In addition, the two isolates had multiple amino acid changes in nucleo-capsid protein, fusion protein, and hemagglutinin/neuraminidase, major antigenic proteins. This allows the further recognition of the presence of BPIV3 type B in Chinese cattle herds. We hope this will help trace the origin of BPIV3, improve the understanding of differences between genotypes, and provide data support for vaccine development.

**Keywords:** bovine parainfluenza virus type 3; genetic phylogenetic analysis; virus identification; indirect immunofluorescence; virus isolation; next-generation sequencing

## 1. Introduction

Bovine parainfluenza virus 3 (BPIV3) is one of several viruses that contribute to bovine respiratory disease complex (BRDC). Under stressful conditions, cattle are highly susceptible to BPIV3 infection, which weakens their immunity and leads to secondary bacterial and mycoplasma infections, eventually causing BRDC [1]. Additionally, it has been reported that BPIV3 shows cross-species transmission, including humans [2] and sheep [3]. BPIV3 was first reported in the United States in 1959 [4], where it was first isolated. Since then, it has been reported in many countries worldwide. In Canada, Peter et al. [5] tested 1745 serum samples from 295 farms for BPIV3 antibodies using an enzymelinked immunosorbent assay and found a positivity rate of 93.3%. In Turkey, Muftuoglu et al. [6] tested 1307 serum samples randomly collected from 2015 to 2019 using a standard virus neutralization test and found a positivity rate of 56.2%. Newcomer et al. [7] performed neutralization tests of three BPIV3 genotypes using sera from five unimmunized herds in the United States. They found that antibody titers were significantly higher for genotype B than for genotypes A and C. In China, Wang et al. [8] tested 2489 bovine serum samples from 12 provinces for BPIV3 antibodies and found a positivity rate of 77%, indicating that BPIV3 is widespread in China.



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