

Full-Genome Sequence of Chicken Anemia Virus Strain GXC060821, Isolated from a Guangxi Sanhuang Chicken

Zhixun Xie, Xianwen Deng, Liji Xie, Jiabo Liu, Yaoshan Pang, Zhiqin Xie, Qing Fan, Sisi Luo

Guangxi Key Laboratory of Animal Vaccines and Diagnostics, Guangxi Veterinary Research Institute, Nanning, Guangxi Province, China

We report here the complete genomic sequence of a novel chicken anemia virus strain GXC060821, isolated from a Sanhuang chicken in Guangxi Province of southern China. The complete genome of GXC060821 was sequenced. The full-length of GXC060821 is 2,292 bp and contains three overlapping open reading frames (ORFs). A comparison of the complete sequences and the deduced amino acid sequences of GXC060821 with 31 other published chicken anemia virus sequences showed that the homologies of the nucleotides are 96.1% to 98.5% and the homologies of the deduced amino acid sequences are 89.8% to 94.2%. Phylogenetic tree analysis indicated that GXC060821 is closely related to the two Chinese strains, TJBD40 (accession no. AY843527) and LF4 (accession no. AY839944), and it has a distant relationship with the American isolate 98D06073 (accession no. AF311900). This report will help to understand the epidemiology and molecular characteristics of chicken anemia virus in a Guangxi Sanhuang chicken.

Received 15 January 2014 Accepted 23 January 2014 Published 20 February 2014

Citation Xie Z, Deng X, Xie L, Liu J, Pang Y, Xie Z, Fan Q, Luo S. 2014. Full-genome sequence of chicken anemia virus strain GXC060821, isolated from a Guangxi Sanhuang chicken. Genome Announc. 2(1):e00040-14. doi:10.1128/genomeA.00040-14.

Copyright © 2014 Xie et al. This is an open-access article distributed under the terms of the Creative Commons Attribution 3.0 Unported license.

Address correspondence to Zhixun Xie, xiezhixun@126.com, or Liji Xie, xie3120371@126.com.

hicken anemia virus (CAV) is a small DNA virus with a circular, covalently linked, single negative-strand genome. It is the causative agent of chicken infectious anemia (CIA) and classified in the family *Circoviridae*, genus *Gyrovirus* (1). CAV is an economically important pathogen with a worldwide distribution. CAV was first isolated in 1979 in Japan and is the major agent responsible for a disease causing severe anemia and immunosuppression (2). The characteristic symptoms of the disease include aplasia of the bone marrow and the destruction of T-lymphoid tissue, which has been shown histopathologically after CAV infection (3, 4). Generally, CAV as the causative agent of chicken anemia disease affects 1-day-old chicks that lack maternal antibodies (5). Mortality rates as high as 55% and morbidity rates as high as 80% have been described when chicks are infected with CAV (6, 7).

In this study, a novel strain of CAV, named GXC060821, was first isolated from a Sanhuang chicken in Guangxi, southern China. The nucleotide sequences of this strain were amplified by PCR. The amplified products were purified and cloned into the pMD-18T vector (TaKaRa, Dalian, China) and then sequenced (Invitrogen, Guangzhou, China) (8). The sequences were assembled and manually edited to generate the final genome sequence.

Sequence analysis showed that the full-genome sequence of GXC060821 is 2,292 nucleotides and contains 3 overlapping open reading frames (ORFs), including viral protein 1 (VP1), viral protein 2 (VP2), and viral protein 3 (VP3) (7–9). The full lengths of these ORFs are 1,350, 651, and 366 nucleotides, respectively.

GXC060821 was compared with 31 CAV stains, four American strains, one Australian strain, one German strain, two Japanese strains, six Malaysian strains, 10 Chinese strains, and seven strains from the United Kingdom. The nucleotide sequence identities of the VP1, VP2, and VP3 genes between GXC060821 and 31 CAV strains are 94.4% to 98.6%, 98.6% to 99.5%, and 98.6% to 99.7%,

respectively, and the amino acid sequence identities are 96.7% to 99.6%, 96.8% to 98.6%, and 96.7% to 99.2%, respectively.

Immunogenicity studies have shown that VP1 and VP2 are crucial components for the elicitation of host-produced virus neutralizing antibodies in chickens (10), and the amino acid sequence of GXC060821 with other 31 CAV strains is conservative. Therefore, VP1 and VP2 have previously been thought to be good candidates for use as immunogens when developing subunit vaccines or diagnostic kits (10, 11).

The complete sequences and the deduced amino acid sequences of GXC060821 with 31 other published CAV sequences showed that the homologies of the nucleotides are 96.1% to 98.5% and the homologies of the deduced amino acid sequences are 89.8% to 94.2%. Phylogenetic tree analysis indicated that GXC060821 is closely related to the two Chinese strains, TJBD40 (GenBank accession no. AY843527) and LF4 (GenBank accession no. AY839944), and it has a distant relationship with the American isolate 98D06073 (GenBank accession no. AF311900). This report will help to understand the epidemiology and molecular characteristics of CAV in a Guangxi Sanhuang chicken.

Nucleotide sequence accession number. The complete genomic sequence of GXC060821 was deposited in GenBank under the accession no. JX964755.

ACKNOWLEDGMENTS

This work was supported by the Guangxi Science and Technology Bureau (grant no. 1222003-2-4) and by the Guangxi Government Senior Scientist Foundation (grant no.2011B020).

REFERENCES

 Pringle CR. 1999. Virus taxonomy at the XIth International Congress of Virology, Sydney, Australia, 1999. Arch. Virol. 144:2065–2069.

- 2. Yuasa N, Taniguchi T, Yoshida I. 1979. Isolation and some characteristics of an agent inducing anemia in chicks. Avian Dis. 23:366–385. http://dx.doi.org/10.2307/1589567.
- 3. Yuasa N, Imai K, Watanabe K, Saito F, Abe M, Komi K. 1987. Aetiological examination of an outbreak of haemorrhagic syndrome in a broiler flock in Japan. Avian Pathol. 16:521–526. http://dx.doi.org/10.1080/03079458708436401.
- Lucio B, Schat KA, Shivaprasad HL. 1990. Identification of the chicken anemia agent, reproduction of the disease, and serological survey in the United States. Avian Dis. 34:146–153. http://dx.doi.org/10.2307/1591346.
- Yuasa N, Noguchi T, Furuta K, Yoshida I. 1980. Maternal antibody and its effect on the susceptibility of chicks to chicken anemia agent. Avian Dis. 24:197–201. http://dx.doi.org/10.2307/1589779.
- Lee MS, Chou YM, Lien YY, Lin MK, Chang WT, Lee HZ, Lee MS, Lai GH, Chen HJ, Huang CH, Lin WH. 2011. Production and diagnostic application of a purified, E. coli-expressed, serological-specific chicken anaemia virus antigen VP3. Transbound. Emerg. Dis. 58:232–239.
- 7. Lai GH, Lin MK, Lien YY, Fu JH, Chen HJ, Huang CH, Tzen J, Lee MS. 2013. Expression and characterization of highly antigenic domains of

- chicken anemia virus viral VP2 and VP3 subunit proteins in a recombinant *E. coli* for sero-diagnostic applications. BMC Vet. Res. 9:161. http://dx.doi.org/10.1186/1746-6148-9-161.
- Xie Z, Xie L, Pang Y, Lu Z, Xie Z, Sun J, Deng X, Liu J, Tang X, Khan M. 2008. Development of a real-time multiplex PCR assay for detection of viral pathogens of penaeid shrimp. Arch. Virol. 153:2245–2251. http://dx.doi.org/10.1007/s00705-008-0253-0.
- 9. Zhang X, Liu Y, Wu B, Sun B, Chen F, Ji J, Ma J, Xie Q. 2013. Phylogenetic and molecular characterization of chicken anemia virus in southern China from 2011 to 2012. Sci. Rep. 3:3519. http://dx.doi.org/10.1038/srep03519.
- Noteborn MH, Verschueren CA, Koch G, Van der Eb AJ. 1998. Simultaneous expression of recombinant baculovirus-encoded chicken anemia virus (CAV) proteins VP1 and VP2 is required for formation of the CAV-specific neutralizing epitope. J. Gen. Virol. 79(Pt 12):3073–3077.
- Koch G, van Roozelaar DJ, Verschueren CA, van der Eb AJ, Noteborn MHM. 1995. Immunogenic and protective properties of chicken anemia virus proteins expressed by baculovirus. Vaccine 13:763–770. http://dx .doi.org/10.1016/0264-410X(94)00034-K.