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Complete Genome Sequence of a Novel Newcastle Disease Virus Strain Isolated from a Chicken in West Africa

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The complete genome sequence of an African Newcastle disease virus (NDV) strain isolated from a chicken in Togo in 2009 was determined. The genome is 15,198 nucleotides (nt) in length and is classified in genotype VII in the class II cluster. Compared to common vaccine strains, the African strain contains a previously described 6-nt insert in the downstream untranslated region of the N gene and a novel 6-nt insert in the HN-L intergenic region. Genome length differences are a marker of the natural history of NDV. This is the first description of a class II NDV strain with a genome of 15,198 nt and a 6-nt insert in the HN-L intergenic region. Sequence divergence relative to vaccine strains was substantial, likely contributes to outbreaks, and illustrates the continued evolution of new NDV strains in West Africa.

Newcastle disease virus (NDV) causes a devastating disease in chickens worldwide (2). NDV is a member of the family *Paramyxoviridae* and has a nonsegmented, negative-sense RNA genome consisting of six transcriptional units (3′-NP-P-M-F-HN-L-5′) (8). NDV strains have been classified into classes I (9 genotypes) and II (11 genotypes) (4). Class I strains are usually avirulent and have genome lengths of 15,198 nucleotides (nt). Class II contains both virulent and avirulent strains. Class II strains isolated prior to 1960 (genotypes I to IV and IX) have genomes of 15,186 nt, while more recent strains (genotypes V to VIII and XI) have genomes of 15,192 nt. Relative to the 15,186-nt genome, the 15,192-nt genome has an insertion of 6 nt in the 5′ (downstream) noncoding region of the N gene and the 15,198-nt genome has an insertion of 12 nt in the P open reading frame (ORF) that increases the length of P by four amino acids. Thus, NDV is a diverse and continually evolving virus (1, 5, 6). Predominantly circulating strains associated with disease outbreaks are genotypes V to VIII of class II (9).

In West Africa, the increasing reports of NDV outbreaks in vaccinated flocks suggest the emergence of new NDV strains that may be somewhat mismatched with common vaccine strains (10). However, little sequence information is available for NDV strains circulating in this region, and molecular characterization of NDV strains has involved partial sequences of the F gene (3). To date, no complete genome sequence of African NDV strains has been available. In this study, the complete consensus genome sequence of an African NDV strain (chicken/Togo/AKO18/2009) isolated from a live bird market in Togo was determined by reverse transcription-PCR using overlapping consensus primers and direct sequencing. The 3′ and 5′ termini were determined by rapid amplification of cDNA ends (11). The genome was found to be 15,198 nt in length and contains the above-mentioned insert in the N gene as well as a novel 6-nt insert in the intergenic sequence between the HN and L genes (^{8338}-UUUUUU-^{8343}), compared to the 15,186-nt genomes of vaccine strains B1 and LaSota (class II, genotype II). Phylogenetic analysis classified this African strain into class II, genotype VII. The amino acid sequence identities of the N, P, M, F, HN, and L proteins between the African strain and the strains LaSota and B1 (which share 99% identity) are 91%, 82%, 89%, 89%, 87%, and 93%, respectively. This indicates that the African strain is substantially distinct from the vaccine strains in use, suggesting that antigenic differences might affect the efficacy of current vaccines and vaccination practice.

The sequence of the F protein cleavage site is a major determinant of NDV pathogenicity (7). The cleavage sites of virulent NDV strains contain multiple basic residues, whereas avirulent strains have fewer basic residues. The African strain has a virulent F protein cleavage-site sequence (RRRKR ↓ F). The data presented here provide evidence of a novel strain with a novel genome length variation circulating in West Africa.

Nucleotide sequence accession number. The genome sequence of the NDV strain has been deposited in GenBank (accession number JX390609).

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REFERENCES

1. **Aldous EW, Fuller CM, Mynn JK, Alexander DJ.** 2004. A molecular epidemiological investigation of isolates of the variant avian paramyxovirus type 1 virus (PPMV-1) responsible for the 1978 to present panzootic in pigeons. Avian Pathol. **33**:258–269.
2. **Alexander DJ.** 1989. Newcastle disease, p 114–120. *In* Purchase HG, Arp LH, Domermuth CH, Pearson JE (ed), A laboratory manual for the isolation and identification of avian pathogens, 3rd ed. American Association of Avian Pathologists, Kendall/Hunt Publishing Company, Dubuque, IA.
3. **Cattoli G, et al.** 2010. Emergence of a new genetic lineage of Newcastle disease virus in West and Central Africa—implications for diagnosis and control. Vet. Microbiol. **142**:168–176.
4. **Czeglédi A, et al.** 2006. Third genome size category of avian paramyxovirus serotype 1 (Newcastle disease virus) and evolutionary implications. Virus Res. **120**:36–48.
5. **Krishnamurthy S, Samal SK.** 1998. Nucleotide sequence of the trailer, nucleocapsid protein gene and intergenic regions of Newcastle disease virus strain Beaudette C and completion of the entire genome sequence. J. Gen. Virol. **79**:2419–2424.
6. **Miller PJ, Decanini EL, Afonso CL.** 2010. Newcastle disease: evolution of genotypes and the related diagnostic challenges. Infect. Genet. Evol. **10**:26–35.
7. **Panda A, Huang Z, Elankumaran S, Rockemann DD, Samal SK.** 2004. Role of fusion protein cleavage site in the virulence of Newcastle disease virus. Microb. Pathog. **36**:1–10.
8. **Samal SK.** 2011. Newcastle disease and related avian paramyxoviruses, p 69–114. *In* Samal SK (ed), The biology of paramyxoviruses. Caister Academic Press, Norfolk, United Kingdom.
9. **Snoeck CJ, et al.** 2009. Newcastle disease virus in West Africa: new virulent strains identified in non-commercial farms. Arch. Virol. **154**:47–54.
10. **Solomon P, Abolnik C, Joannis TM, Bisschop S.** 2012. Virulent Newcastle disease virus in Nigeria: identification of a new clade of sub-lineage 5f from livebird markets. Virus Genes **44**:98–103.
11. **Xiao S, et al.** 2012. Complete genome sequences of Newcastle disease virus strains circulating in chicken populations of Indonesia. J. Virol. **86**: 5969–5970.