AMERICAN SOCIETY FOR MICROBIOLOGY | Microbiology Resource Announcements

GENOME SEQUENCES

Near-Full-Length Genome Sequence of a Foot-and-Mouth Disease Virus of Serotype Southern African Territories 2 Isolated from Nigeria in 2014

Ian H. Fish,[a,b] David Vierra,[a,c] David O. Ehizibolo,[d] Rachel Palinski,[a] Miranda R. Bertram,[a,b] Steven J. Pauszek,[a] Ethan J. Hartwig,[a] George R. Smoliga,[a] Jonathan Arzt[a]

[a]Foreign Animal Disease Research Unit, Plum Island Animal Disease Center, ARS-USDA, Orient Point, New York, USA
[b]Oak Ridge Institute for Science and Education, PIADC Research Participation Program, Oak Ridge, Tennessee, USA
[c]Department of Diagnostic Medicine and Pathobiology, College of Veterinary Medicine, Kansas State University, Manhattan, Kansas, USA
[d]Virology Division, National Veterinary Research Institute, Vom, Nigeria

ABSTRACT We report a near-full-length genome sequence of a foot-and-mouth disease virus (FMDV) of serotype Southern African Territories 2 (SAT 2), topotype VII, isolated from cattle during an FMDV outbreak in Bauchi State, Nigeria, in October 2014. This provides the first SAT 2 near-full-length genome sequence from West Africa and contributes to our understanding of viral spread and evolution.

Foot-and-mouth disease virus (FMDV; family *Picornaviridae*, genus *Aphthovirus*), the etiologic agent of foot-and-mouth disease, is one of the most economically impactful infectious diseases of livestock. Acutely infected animals develop characteristic vesicular lesions on the feet, tongue, snout, and teats (1, 2). The seven distinct FMDV serotypes (A, Asia 1, C, O, and Southern African Territories [SAT] 1 to -3) are each defined based on VP1 coding sequences (3). SAT 2 was first identified in West Africa in 1974 (4) and is subdivided into 14 topotypes (https://www.wrlfmd.org/fmdv-genome/fmd-prototype-strains#panel-8976). Topotype VII was first identified in Nigeria in 2007 (SAT2/NIG/2/07) (5). Transboundary livestock movement in West Africa contributes to FMDV spread (6), and knowledge of the endemicity of FMDV genotypes is essential to understanding its molecular epidemiology. These considerations are important components of the Progressive Control Pathway for FMDV (PCP-FMDV) in Nigeria (7).

The FMDV genome reported herein was isolated from cattle epithelium during an FMDV outbreak in Magama, Bauchi, Nigeria, in October 2014. The sample tested positive for FMDV by virus isolation on LFBK-$\alpha_v\beta_6$ cells and was confirmed with FMDV-specific reverse transcriptase quantitative PCR (qRT-PCR) using the primers 5′-GACAAAGGTTTTGTTCTTGGTCA and 5′-TGCGAGTCCTGCCACGGA (8). The total cell supernatant RNA was sequenced as previously described (9, 10). In brief, the virus was passaged once in LFBK-$\alpha_v\beta_6$ cells, and RNA was extracted from the supernatant using the MagMAX total RNA isolation kit, with the host DNA depleted using the DNA-free DNase kit (Ambion). The treated RNA underwent first-strand synthesis using a SuperScript first-strand synthesis system (Invitrogen) with random hexameric primers, a poly(T) primer targeting the 3′ untranslated region (UTR), and one FMDV-specific primer targeting a conserved sequence in coding region 2A. Double-stranded cDNA was generated using a NEBNext Ultra II nondirectional RNA second-strand synthesis module (New England BioLabs) and purified with SPRIselect beads (Beckman Coulter). The sequencing library was constructed using the Nextera XT kit (Illumina) and sequenced on the NextSeq 500 platform. Reads were trimmed and filtered for quality and specific primers. In CLC Genomics Workbench 11.0, consensus sequences were compiled by both *de novo* assembly and read mapping to the full-length reference genome

Citation Fish IH, Vierra D, Ehizibolo DO, Palinski R, Bertram MR, Pauszek SJ, Hartwig EJ, Smoliga GR, Arzt J. 2019. Near-full-length genome sequence of a foot-and-mouth disease virus of serotype Southern African Territories 2 isolated from Nigeria in 2014. Microbiol Resour Announc 8:e00788-19. https://doi.org/10.1128/MRA.00788-19.

Editor Kenneth M. Stedman, Portland State University

Address correspondence to Jonathan Arzt, jonathan.arzt@ars.usda.gov.

Received 16 July 2019
Accepted 8 August 2019
Published 29 August 2019

SAT 2 EGY/9/2012 (GenBank accession number JX014255), with a total of 11,916 reads covering 8,091 nucleotides (nt) in the final assembly and an average depth of 207.1 reads per site.

This 8,091-nt near-full-length genome includes 999 nt of the 5′ UTR, followed by the 7,008-nt polyprotein coding sequence and 84 nt of the 3′ UTR (GC content, 54.4%). Phylogenetic analysis of the VP1 coding sequence identified it as serotype SAT 2, topotype VII, most closely related to viruses detected in Cameroon 3 months following (GenBank accession number MG873224), sharing 99.1% pairwise nucleotide identity (D. O. Ehizibolo and J. Arzt, unpublished data). A BLASTn query of the entire sequence did not identify any sequence with an identity above 80%. This discrepancy is the result of the very limited number of publicly available full-length FMDV genomes.

Given the ongoing efforts to control and eradicate FMDV in West Africa, there is a critical need for contemporary FMDV sequences. The genome sequence detailed in this announcement, SAT2/NIG/1/14, contributes to an understanding of the molecular epidemiology of FMDV in West Africa as part of the pathway to regional control.

Data availability. The assembled sequencing reads of SAT2/NIG/1/14 have been deposited in the NCBI under BioProject accession number PRJNA549649 and the consensus sequence under GenBank accession number MN103523.

ACKNOWLEDGMENTS

This research was funded in part by ARS CRIS project number 1940-32000-061-00D. M. R. Bertram and I. H. Fish were the recipients of a Plum Island Animal Disease Center Research Participation Program fellowship, administered by the Oak Ridge Institute for Science and Education (ORISE) through an interagency agreement between the U.S. Department of Energy (DOE) and the U.S. Department of Agriculture (USDA). All opinions expressed in this paper are the authors' and do not necessarily reflect the policies and views of the USDA, DOE, or ORISE.

REFERENCES

1. Arzt J, Juleff N, Zhang Z, Rodriguez LL. 2011. The pathogenesis of foot-and-mouth disease I: viral pathways in cattle. Transbound Emerg Dis 58:291–304. https://doi.org/10.1111/j.1865-1682.2011.01204.x.
2. Arzt J, Baxt B, Grubman MJ, Jackson T, Juleff N, Rhyan J, Rieder E, Waters R, Rodriguez LL. 2011. The pathogenesis of foot-and-mouth disease II: viral pathways in swine, small ruminants, and wildlife; myotropism, chronic syndromes, and molecular virus-host interactions. Transbound Emerg Dis 58:305–326. https://doi.org/10.1111/j.1865-1682.2011.01236.x.
3. Knowles NJ, Samuel AR. 2003. Molecular epidemiology of foot-and-mouth disease virus. Virus Res 91:65–80. https://doi.org/10.1016/S0168-1702(02)00260-5.
4. Valdazo-González B, Knowles NJ, Hammond J, King DP. 2012. Genome sequences of SAT 2 foot-and-mouth disease viruses from Egypt and Palestinian Autonomous Territories (Gaza Strip). J Virol 86:8901–8902. https://doi.org/10.1128/JVI.01231-12.
5. Ehizibolo DO, Haegeman A, De Vleeschauwer AR, Umoh JU, Kazeem HM, Okolocha EC, Van Borm S, De Clercq K. 2017. Detection and molecular characterization of foot and mouth disease viruses from outbreaks in some states of Northern Nigeria 2013–2015. Transbound Emerg Dis 64:1979–1990. https://doi.org/10.1111/tbed.12602.
6. Tekleghiorghis T, Moormann RJ, Weerdmeester K, Dekker A. 2016. Foot-and-mouth disease transmission in Africa: implications for control, a review. Transbound Emerg Dis 63:136–151. https://doi.org/10.1111/tbed.12248.
7. Bronsvoort BM, Radford AD, Tanya VN, Nfon C, Kitching RP, Morgan KL. 2004. Molecular epidemiology of foot-and-mouth disease viruses in the Adamawa Province of Cameroon. J Clin Microbiol 42:2186–2196. https://doi.org/10.1128/JCM.42.5.2186-2196.2004.
8. Stenfeldt C, Pacheco JM, Smoliga GR, Bishop E, Pauszek SJ, Hartwig EJ, Rodriguez LL, Arzt J. 2016. Detection of foot-and-mouth disease virus RNA and capsid protein in lymphoid tissues of convalescent pigs does not indicate existence of a carrier state. Transbound Emerg Dis 63:152–164. https://doi.org/10.1111/tbed.12235.
9. Palinski RM, Bertram MR, Vu LT, Pauszek SJ, Hartwig EJ, Smoliga GR, Stenfeldt C, Fish IH, Hoang BH, Phuong NT, Hung VV, Vu PP, Dung NK, Dong PV, Tien NN, Tho ND, Dung DH, Arzt J. 2019. First genome sequence of foot-and-mouth disease virus serotype O sublineage Ind2001e from southern Vietnam. Microbiol Resour Announc 8:e01424-18. https://doi.org/10.1128/MRA.01424-18.
10. Logan G, Freimanis GL, King DJ, Valdazo-Gonzalez B, Bachanek-Bankowska K, Sanderson ND, Knowles NJ, King DP, Cottam EM. 2014. A universal protocol to generate consensus level genome sequences for foot-and-mouth disease virus and other positive-sense polyadenylated RNA viruses using the Illumina MiSeq. BMC Genomics 15:828. https://doi.org/10.1186/1471-2164-15-828.