

VIRUSES



Genome Sequence of a Bovine Rhinitis B Virus Identified in Cattle in Sweden

gen@meAnnouncements™

^(D) Anne-Lie Blomström,^a Veslemøy Oma,^b Mamata Khatri,^c Hanne H. Hansen,^d Maria Stokstad,^b Mikael Berg,^a Mette Myrmel^c

Department of Biomedical Sciences and Veterinary Public Health, Section of Virology, Swedish University of Agricultural Sciences, Uppsala, Sweden^a; Department of Production Animal Clinical Sciences, Faculty of Veterinary Medicine, Norwegian University of Life Sciences, Oslo, Norway^b; Department for Food Safety and Infection Biology, Faculty of Veterinary Medicine, Norwegian University of Life Sciences, Oslo, Norway^c; Faculty of Biosciences, Center for Integrative Genetics (CIGENE), Norwegian University of Life Sciences, Ås, Norway^d

ABSTRACT A bovine rhinitis B virus, identified in a calf from Sweden, was genetically characterized. The complete polyprotein was recovered, and phylogenetic analysis showed that this virus has the highest similarity to a bovine rhinitis B virus previously identified in Mexico.

Bindustry worldwide. Several viruses have been associated with BRD, such as bovine coronavirus (BCoV), bovine respiratory syncytial virus, bovine parainfluenza virus 3, and bovine herpesvirus type 1 (1, 2). In a sample from an experimental study investigating BCoV (3), a nearly full-length genome of a bovine rhinitis B virus (BRBV) was identified.

A nasal swab was collected from a BcoV-infected calf in Sweden, and the sample was treated with RNase A and DNase prior to RNA extraction. The RNA was reverse transcribed and amplified using the Ovation RNA sequencing (RNA-Seq) system version 2, according to the manufacturer's instructions (two technical replicates). The two amplified samples were prepared for sequencing using the Illumina Nextera XT DNA library preparation kit, and they were sequenced on the MiSeq platform. Between six and nine million reads were obtained per sample. The raw data from the MiSeq run were quality checked and trimmed ($Q \ge 30$; maximum number of ambiguities = 2) using CLC Genomics Workbench in order to remove poor data. The reads were mapped to BcoV, and the remaining unmapped reads were de novo assembled using CLC Genomics Workbench and subjected to blastx searches using Diamond (4). Apart from BCoV and a low number of phage and retrovirus sequences, another positive-sense single-stranded RNA (ssRNA) virus, BRBV, was recognized. About 9,000 and 11,000 BRBV reads were identified in the 2 data sets. The BRBV reads were extracted and de novo creating a contig of 5,760 nucleotides that mapped to the BRBV genome. In order to recover a larger portion of the genome, the contig and all the trimmed raw reads were mapped against both BRBV strain Mexb10 (GenBank accession no. KU159357.1) and TCH5 (GenBank accession no. KU168861.1). This rendered the nearly complete BRBV genome, including the entire sequence coding for the polyprotein (GenBank accession no. KY432299). MEGA6 (5) was used for ClustalW alignment and phylogeny (using the neighbor-joining method and a bootstrap value of 1,000) of the identified BRBV and the other 9 nearly complete BRBV genomes available in GenBank. The BRBV identified in this study (BRBV_SWE1) clustered together with a BRBV strain (Mexb10) collected from beef cattle in Mexico in 2015 (6). The nucleotide sequence similarity between the two sequences was 83.8%. The nucleotide similarity between BRBV_SWE1 and other BRBV strains varied between 71 and 75%. On an amino acid level, the similarity varied between 85 and 94%. Looking at the sequence resemblance across the genome, VP2, VP3, and VP1 had the highest variation. The importance of BRBV as a pathogen in cattle

Received 14 February 2017 Accepted 20 March 2017 Published 11 May 2017

Citation Blomström A-L, Oma V, Khatri M, Hansen HH, Stokstad M, Berg M, Myrmel M. 2017. Genome sequence of a bovine rhinitis B virus identified in cattle in Sweden. Genome Announc 5:e00172-17. https://doi.org/10.1128/ genomeA.00172-17.

Copyright © 2017 Blomström et al. This is an open-access article distributed under the terms of the Creative Commons Attribution 4.0 International license.

Address correspondence to Anne-Lie Blomström, anne-lie.blomstrom@slu.se. is currently unknown, but BRBV has been identified in several other metagenomic studies investigating BRD (6–8).

Accession number(s). The nucleotide sequence of BRBV_SWE1 has been deposited in GenBank under the accession no. KY432299.

ACKNOWLEDGMENTS

This project was funded by the Research Council of Norway (NFR-project no. 224771/E40), the Norwegian Research Funding for Agriculture and Food Industry, and TINE Norwegian Dairies BA. Financial support was also partly from the Swedish Research Council for Environment, Agricultural Sciences and Spatial Planning, Formas (Formas-2012-586). The blast analysis computations were performed on resources provided by SNIC through Uppsala Multidisciplinary Center for Advanced Computational Science (UPPMAX) under project b2012218.

REFERENCES

- Edwards TA. 2010. Control methods for bovine respiratory disease for feedlot cattle. Vet Clin North Am Food Anim Pract 26:273–284. https:// doi.org/10.1016/j.cvfa.2010.03.005.
- Taylor JD, Fulton RW, Lehenbauer TW, Step DL, Confer AW. 2010. The epidemiology of bovine respiratory disease: what is the evidence for predisposing factors? Can Vet J 51:1095–1102.
- Oma VS, Tråven M, Alenius S, Myrmel M, Stokstad M. 2016. Bovine coronavirus in naturally and experimentally exposed calves; viral shedding and the potential for transmission. Virol J 13:100. https://doi.org/10 .1186/s12985-016-0555-x.
- Buchfink B, Xie C, Huson DH. 2015. Fast and sensitive protein alignment using DIAMOND. Nat Methods 12:59–60. https://doi.org/10.1038/nmeth .3176.
- 5. Tamura K, Stecher G, Peterson D, Filipski A, Kumar S. 2013. MEGA6:

molecular evolutionary genetics analysis version 6.0. Mol Biol Evol 30: 2725–2729. https://doi.org/10.1093/molbev/mst197.

- Mitra N, Cernicchiaro N, Torres S, Li F, Hause BM. 2016. Metagenomic characterization of the virome associated with bovine respiratory disease in feedlot cattle identified novel viruses and suggests an etiologic role for influenza D virus. J Gen Virol 97:1771–1784. https://doi.org/10.1099/jgv.0 .000492.
- Ng TF, Kondov NO, Deng X, Van Eenennaam A, Neibergs HL, Delwart E. 2015. A metagenomics and case-control study to identify viruses associated with bovine respiratory disease. J Virol 89:5340–5349. https://doi .org/10.1128/JVI.00064-15.
- Hause BM, Collin EA, Anderson J, Hesse RA, Anderson G. 2015. Bovine rhinitis viruses are common in U.S. cattle with bovine respiratory disease. PLoS One 10:e0121998. https://doi.org/10.1371/journal.pone.0121998.