



## Genome Sequence of a Bornavirus Recovered from an African Garter Snake (*Elapsoidea loveridgei*)

## Mark D. Stenglein,<sup>a\*</sup> Eli B. Leavitt,<sup>a</sup> Michael A. Abramovitch,<sup>a</sup> Jimmy A. McGuire,<sup>b</sup> Joseph L. DeRisi<sup>a,c</sup>

Department of Biochemistry and Biophysics, University of California San Francisco, San Francisco, California, USA<sup>a</sup>; Department of Integrative Biology and Museum of Vertebrate Zoology, University of California, Berkeley, Berkeley, California, USA<sup>b</sup>; Howard Hughes Medical Institute, Chevy Chase, Maryland, USA<sup>c</sup>

\* Present address: Mark D. Stenglein, Department of Microbiology, Immunology and Pathology, Colorado State University, Fort Collins, Colorado, USA.

M.D.S., E.B.L., M.A.A., and J.A.M. contributed equally to this work

Bornaviruses are known to infect mammals and birds, and they have been associated with disease in both groups of animals. Here, we report the genome sequence of a bornavirus identified in a wild-caught Loveridge's garter snake (*Elapsoidea lover-idgei*).

Received 22 July 2014 Accepted 8 September 2014 Published 9 October 2014

Citation Stenglein MD, Leavitt EB, Abramovitch MA, McGuire JA, DeRisi JL. 2014. Genome sequence of a bornavirus recovered from an African garter snake (*Elapsoidea loveridgei*). Genome Announc. 2(5):e00779-14. doi:10.1128/genomeA.00779-14.

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

Address correspondence to Joseph L. DeRisi, joe@derisilab.ucsf.edu.

The field of reptile virology is significantly understudied relative to that of viruses that infect mammalian species. However, the field is developing quickly, thanks to growing interest and the advent of newer technologies for detecting novel viruses. Many representatives of well-studied viral families have been characterized in reptile species (1–3), and more recently, deep-sequencing approaches have identified entire new clades of viruses from families previously not known to infect reptiles (4, 5).

Bornaviruses (family *Bornaviridae*) are negative-strand RNA viruses that have been associated with disease in horses and, more recently, with proventricular dilatation disease (PDD) in parrots and other psittacines (6, 7). In 2010, fragments containing the X and phosphoprotein (P) genes of a putative bornavirus were recognized in a cDNA library prepared from Gaboon viper (*Bitis gabonica*) RNA (8, 9). To date, a complete genome of a reptile bornavirus (RBV) has yet to be elucidated.

Through a deep-sequencing survey of reptile samples housed in the Field Museum of Natural History, we discovered large numbers of sequences with similarity to those of avian bornaviruses originating from the tissues of an *Elapsoidea loveridgei* (Loveridge's garter snake), specimen FMNH 251327. Initial short contigs were generated with Trinity (10), followed by iterative contig extension using the *de novo* assembler PRICE (11). The resulting assembly consisted of a single 8,884-nucleotide contig. The annotation of the sequence revealed a canonical gene repertoire (N, X, P, M, G, and L) and configuration for bornaviruses, including sequence motifs, such as transcription initiation sites, transcriptional terminators, and a splice donor site for the intron present in L (polymerase). The remapping of the individual sequencing reads revealed a mean coverage of 200-fold ( $\pm$ 91).

A comparison of the nucleotide sequence to that of other bornaviruses revealed that this virus is related to the avian bornaviruses. For example, avian Borna disease virus (isolate bil, Gen-Bank accession no. EU781967) shares 62% pairwise global nucleotide identity. Interestingly, the alignment of the putative P protein sequence with the previously reported RBV P is only 64% identical at the amino acid level, suggesting the existence of a large diverse family of bornaviruses present in reptiles, although no disease association has been recognized to date.

Based on alignment to avian Borna disease virus, we believe this assembly represents the complete genome (12) and thus the first bornavirus genome sequenced from a nonavian reptile.

Nucleotide sequence accession number. The genome sequence has been deposited in GenBank under accession no. KM114265.

## ACKNOWLEDGMENTS

We thank Alan Resetar and the Amphibian and Reptile Collection of the Field Museum of Natural History for the loan of frozen tissue specimens (loan Z-38759).

This research was supported by the Howard Hughes Medical Institute (to J.L.D.) and by National Science Foundation grants DEB 0330750, 0543556, and 1146491 (to J.A.M.).

## REFERENCES

- 1. Marschang RE. 2011. Viruses infecting reptiles. Viruses 3:2087–2126. http://dx.doi.org/10.3390/v3112087.
- Latney LV, Wellehan J. 2013. Selected emerging infectious diseases of Squamata. Vet. Clin. North Am. Exot. Anim. Pract. 16:319–338. http:// dx.doi.org/10.1016/j.cvex.2013.01.003.
- 3. Jacobson ER. 2007. Infectious diseases and pathology of reptiles: color atlas and text. CRC Press/Taylor & Francis, Boca Raton, FL.
- 4. Stenglein MD, Sanders C, Kistler AL, Ruby JG, Franco JY, Reavill DR, Dunker F, Derisi JL. 2012. Identification, characterization, and *in vitro* culture of highly divergent arenaviruses from boa constrictors and annulated tree boas: candidate etiological agents for snake inclusion body disease. mBio 3(4):e00180-12. http://dx.doi.org/10.1128/mBio.00180-12.
- Hyndman TH, Marschang RE, Wellehan JFX, Jr, Nicholls PK. 2012. Isolation and molecular identification of Sunshine virus, a novel paramyxovirus found in Australian snakes. Infect. Genet. Evol. 12: 1436–1446. http://dx.doi.org/10.1016/j.meegid.2012.04.022.
- 6. Lipkin WI, Briese T. 2007. Bornaviridae, p 1829–1851. *In* Knipe DM, Howley PM (ed), Fields Virology, 5th ed. Lippincott Williams & Wilkins, Philadelphia, PA.

- Kistler AL, Gancz A, Clubb S, Skewes-Cox P, Fischer K, Sorber K, Chiu CY, Lublin A, Mechani S, Farnoushi Y, Greninger A, Wen CC, Karlene SB, Ganem D, Derisi JL. 2008. Recovery of divergent avian bornaviruses from cases of proventricular dilatation disease: identification of a candidate etiologic agent. Virol. J. 5:88. http://dx.doi.org/10.1186/1743-422X-5-88.
- 8. Francischetti IM, My-Pham V, Harrison J, Garfield MK, Ribeiro JM. 2004. *Bitis gabonica* (gaboon viper) snake venom gland: toward a catalog for the full-length transcripts (cDNA) and proteins. Gene 337:55–69. http://dx.doi.org/10.1016/j.gene.2004.03.024.
- Horie M, Honda T, Suzuki Y, Kobayashi Y, Daito T, Oshida T, Ikuta K, Jern P, Gojobori T, Coffin JM, Tomonaga K. 2010. Endogenous nonretroviral RNA virus elements in mammalian genomes. Nature 463: 84–87. http://dx.doi.org/10.1038/nature08695.
- 10. Grabherr MG, Haas BJ, Yassour M, Levin JZ, Thompson DA, Amit I,

Adiconis X, Fan L, Raychowdhury R, Zeng Q, Chen Z, Mauceli E, Hacohen N, Gnirke A, Rhind N, di Palma F, Birren BW, Nusbaum C, Lindblad-Toh K, Friedman N, Regev A. 2011. Full-length transcriptome assembly from RNA-Seq data. Nat. Biotechnol. 29:644–652. http://dx.doi.org/10.1038/nbt.1883.

- Ruby JG, Bellare P, DeRisi JL. 2013. PRICE: software for the targeted assembly of components of (Meta)genomic sequence data. G3 (Bethesda) 3:865–880. http://dx.doi.org/10.1534/g3.113.005967.
- 12. Ladner JT, Beitzel B, Chain PS, Davenport MG, Donaldson EF, Frieman M, Kugelman JR, Kuhn JH, O'Rear J, Sabeti PC, Wentworth DE, Wiley MR, Yu GY, The Threat Characterization Consortium, Sozhamannan S, Bradburne C, Palacios G. 2014. Standards for sequencing viral genomes in the era of high-throughput sequencing. mBio 5(3): e01360-14. http://dx.doi.org/10.1128/mBio.01360-14.