EUKARYOTES

Received 31 May 2017 Accepted 8 June 2017 Published 27 July 2017

Citation Obkura M Fitak RR Wisecaver JH

DeBlasio D, Niazi F, Egholm M, Rounsley SD, Kodira CD, Orbach MJ. 2017. Genome sequence of *Ophidiomyces ophiodiicola*, an

emerging fungal pathogen of snakes. Genome

Announc 5:e00677-17. https://doi.org/10.1128/

open-access article distributed under the terms

Copyright © 2017 Ohkura et al. This is an

of the Creative Commons Attribution 4.0

Address correspondence to Marc J. Orbach,

* Present address: Robert R. Fitak, Department

Department of Biological Sciences, Vanderbilt

DeBlasio, Computational Biology Department, Carnegie Mellon University, Pittsburgh,

Pennsylvania, USA; Faheem Niazi, Bristol Myers

Squibb, Wallingford, Connecticut, USA; Michael

University, Nashville, Tennessee, USA: Dan

Egholm, Biopharmaceuticals Division, Pall Corporation, Port Washington, New York, USA;

Steven D. Rounsley, Genus PLC, DeForest,

Computational Biology, GE Global Research Center, Niskayuna, New York, USA.

Wisconsin, USA; Chinnappa D. Kodira,

of Biology, Duke University, Durham, North Carolina, USA: Jennifer H. Wisecaver,

genomeA.00677-17.

International license

orbachmj@email.arizona.edu.



AMERICAN SOCIETY FOR MICROBIOLOGY

Genome Sequence of *Ophidiomyces ophiodiicola*, an Emerging Fungal Pathogen of Snakes

Mana Ohkura,^a Robert R. Fitak,^{a*} Jennifer H. Wisecaver,^{a*} Dan DeBlasio,^{a*} Faheem Niazi,^{b*} Michael Egholm,^{b*} Steven D. Rounsley,^{a*} Chinnappa D. Kodira,^{b*} Marc J. Orbach^a

gen@meAnnouncements™

School of Plant Sciences, University of Arizona, Tucson, Arizona, USA^a; 454 Life Sciences, a Roche Company, Branford, Connecticut, USA^b

ABSTRACT *Ophidiomyces ophiodiicola*, which belongs to the order *Onygenales*, is an emerging fungal pathogen of snakes in the United States. This study reports the 21.9-Mb genome sequence of an isolate of this reptilian pathogen obtained from a black racer snake in Pennsylvania.

Ophidiomyces ophiodiicola is a keratinophilic fungal pathogen of the order Onygenales and is an emerging threat to wild and captive snakes in the United States (1). Isolates have also been reported in Germany, England, and Australia (2, 3). In the United States, O. ophiodiicola has been described on several snake species in the Northeast, the Southeast, and the Midwest; it is often characterized by skin lesions around the head but is also known to produce invasive infections. This pathogen belongs to the family Onygenaceae, which includes the saprotroph Uncinocarpus reesei and the human pathogens belonging to the genus Coccidioides. To develop an understanding of how pathogenicity on snakes evolved in this fungus, we sequenced and assembled its genome.

The isolate of O. ophiodiicola used for sequencing was recovered from a skeletal lesion of a captive black racer (Coluber constrictor) at the Pennsylvania State Animal Diagnostic Laboratory. Taxonomic analysis of this fungus was performed (4), and the isolate was deposited in the University of Arizona Mycological Herbarium (MYCO-ARIZ AN0400001). Genomic DNA of the isolate was prepared from mycelium grown 4 days in liquid medium, using a liquid N₂ grinding procedure for DNA extraction. Two DNA libraries were prepared for sequencing: one a shotgun library and the other a 3-kb paired-end insert library. Each library was sequenced on one picotiter plate using 454 FLX-Titanium chemistry by Roche (5). The >3.1 million 454 pyrosequencing reads were assembled using Roche's Newbler assembly software. The sequences were assembled into 39 scaffolds for a total genome of 21.9 Mb with a scaffold N_{50} measure of 1.5 Mb and a GC content of 47%. Completeness of the assembly was assessed by the presence of 439 (95.9%) of known core eukaryotic genes using CEGMA (6). Genes were predicted using the MAKER version 2.31 annotation pipeline (7) with AUGUSTUS, SNAP, and GENEMARK-ES as gene callers (8-10). The core eukaryotic genes identified by CEGMA (6) and proteins from Coccidioides immitis RS were provided as protein evidence to train MAKER, and a total of 7,287 genes were predicted.

Accession number(s). This whole-genome shotgun project has been deposited at DDBJ/ENA/GenBank under the accession number MWKM00000000. The version described in this paper is the first version, MWKM01000000.

ACKNOWLEDGMENT

M.O., R.R.F., J.H.W., and D.D. were supported by the NSF IGERT program (grant DGE-0654435).

REFERENCES

- Allender MC, Raudabaugh DB, Gleason FH, Miller AN. 2015. The natural history, ecology, and epidemiology of *Ophidiomyces ophiodiicola* and its potential impact on free-ranging snake populations. Fungal Ecol 17: 187–196. https://doi.org/10.1016/j.funeco.2015.05.003.
- Vissiennon T, Schüppel KF, Ullrich E, Kuijpers AFA. 1999. Case report. A disseminated infection due to *Chrysosporium queenslandicum* in a garter snake (*Thamnophis*). Mycoses 42:107–110. https://doi.org/10.1046/j.1439 -0507.1999.00409.x.
- Sigler L, Hambleton S, Paré JA. 2013. Molecular characterization of reptile pathogens currently known as members of the *Chrysosporium* anamorph of *Nannizziopsis vriesii* complex and relationship with some human-associated isolates. J Clin Microbiol 51:3338–3357. https://doi .org/10.1128/JCM.01465-13.
- Ohkura M, Worley JJ, Hughes-Hallett JE, Fisher JS, Love BC, Arnold AE, Orbach MJ. 2016. Ophidiomyces ophidiicola on a captive black racer (*Coluber constrictor*) and a garter snake (*Thamnophis sirtalis*) in Pennsylvania. J Zoo Wildl Med 47:341–346. https://doi.org/10.1638/2015-0123.1.
- Margulies M, Egholm M, Altman WE, Attiya S, Bader JS, Bemben LA, Berka J, Braverman MS, Chen YJ, Chen ZT, Dewell SB, Du L, Fierro JM, Gomes XV, Godwin BC, He W, Helgesen S, Ho CH, Irzyk GP, Jando SC, Alenquer MLI, Jarvie TP, Jirage KB, Kim JB, Knight JR, Lanza JR, Leamon JH, Lefkowitz SM, Lei M, Li J, Lohman KL, Lu H, Makhijani VB, McDade KE,

McKenna MP, Myers EW, Nickerson E, Nobile JR, Plant R, Puc BP, Ronan MT, Roth GT, Sarkis GJ, Simons JF, Simpson JW, Srinivasan M, Tartaro KR, Tomasz A, Vogt KA, Volkmer GA, Wang SH, Wang Y, Weiner MP, Yu PG, Begley RF, Rothberg JM. 2005. Genome sequencing in microfabricated high-density picolitre reactors. Nature 437:376–380. https://doi.org/10.1038/nature03959.

- Parra G, Bradnam K, Korf I. 2007. CEGMA: a pipeline to accurately annotate core genes in eukaryotic genomes. Bioinformatics 23: 1061–1067. https://doi.org/10.1093/bioinformatics/btm071.
- Holt C, Yandell M. 2011. MAKER2: an annotation pipeline and genomedatabase management tool for second-generation genome projects. BMC Bioinformatics 12:491. https://doi.org/10.1186/1471-2105-12-491.
- Stanke M, Waack S. 2003. Gene prediction with a hidden Markov model and a new intron submodel. Bioinformatics 19:ii215–ii225. https://doi .org/10.1093/bioinformatics/btg1080.
- 9. Korf I. 2004. Gene finding in novel genomes. BMC Bioinformatics 5:59. https://doi.org/10.1186/1471-2105-5-59.
- Ter-Hovhannisyan V, Lomsadze A, Chernoff YO, Borodovsky M. 2008. Gene prediction in novel fungal genomes using an ab initio algorithm with unsupervised training. Genome Res 18:1979–1990. https://doi.org/ 10.1101/gr.081612.108.