



Draft Genome Sequence of *Simplicillium aogashimaense* 72-15.1, a Putative Endophyte of *Brachiaria brizantha*

Ruy Jauregui,^a Linda J. Johnson,^a Suliana E. Teasdale^{a*}

^aAgResearch Limited, Grasslands Research Centre, Palmerston North, New Zealand

Ruy Jauregui and Linda J. Johnson contributed equally to this work. Author order was determined based on bioinformatics contribution.

ABSTRACT Here, we report a 29-Mb draft genome sequence of strain 72-15.1 of *Simplicillium aogashimaense* (Cordycipitaceae, Hypocreales). Strain 72-15.1 was a unique fungal isolate obtained from asymptomatic tillers of the tropical grass *Brachiaria brizantha*.

Simplicillium aogashimaense strain 72-15.1 is a putative fungal endophyte of *Brachiaria brizantha* (host accession number CIAT 16320) and was isolated from an asymptomatic leaf (1). This species was originally described from soil samples in Asia (2). Preliminary identification of *S. aogashimaense* strain 72-15.1 was based on morphological characteristics and a phylogenetic analysis with the internal transcribed spacer (ITS) region (1). *S. aogashimaense* strain 72-15.1 was isolated only once from *B. brizantha* and was the only species of the *Simplicillium* genus (as described by Zare and Gams [3]) to be isolated from nine species of *Brachiaria* grasses (1).

The 29-Mb draft genome of *S. aogashimaense* strain 72-15.1 is 1 of only 13 genomes of *Simplicillium* species to have been described thus far (4, 5). *Simplicillium* species are both ecologically and economically interesting due to their widespread distribution, diverse host specificities, varied lifestyles (4, 5), utility as biocontrol agents (6–8), and production of bioactive compounds (9–13). Previously, we showed high antifungal activity of the sequenced strain 72-15.1 against the phytopathogenic fungi *Alternaria alternatum*, *Bipolaris* sp. aff. *sorokiniana*, and *Curvularia trifolii* (1). The genome of strain 72-15.1 can therefore be mined for the discovery of these antifungal secondary metabolites, as well as for other metabolites that may be responsible for the antimicrobial properties exhibited by some members of this genus against bacterial and fungal plant pathogens as well as plant parasitic nematodes (8, 14, 15).

To obtain fungal material for DNA extraction, a pure culture was lacerated with sterile water, and the resulting solution was spread onto a sheet of cellophane on the surface of a peptone-dextrose agar (PDA) plate and then incubated at 24°C for ~4 days. High-quality DNA was extracted from strain 72-15.1 using a Zymo Research Quick-DNA fungal/bacterial miniprep kit, following the manufacturer's protocol, except mycelium was ground with a plastic pestle fitted to a drill under liquid nitrogen. Two libraries were constructed for whole-genome shotgun sequencing, one for paired-end reads with an insert size of 350 nucleotides (nt) and one for mate pair reads with an insert size of 7 kb, using the Illumina TruSeq DNA Nano low-throughput (LT) 350-bp and Illumina Nextera mate pair gel-plus kits. The libraries were sequenced on an Illumina HiSeq 2000 instrument using v3 chemistry. The instrument produced 30 million paired-end read pairs and 33 million mate pair read pairs with a read length of 125 nt. All paired reads were quality trimmed and filtered using Trimmomatic v0.33 (16) and assembled using Edena v3.2 (17) with default parameters. Contigs shorter than 200 nt were discarded. The contigs were further assembled into scaffolds using the program SSPACE v3.0 (18) with the parameters -k 5, -a 0.7, -x 1, -m 30, and -o 20. The pipeline produced 22

Citation Jauregui R, Johnson LJ, Teasdale SE. 2020. Draft genome sequence of *Simplicillium aogashimaense* 72-15.1, a putative endophyte of *Brachiaria brizantha*. Microbiol Resour Announc 9:e00366-20. <https://doi.org/10.1128/MRA.00366-20>.

Editor Jason E. Stajich, University of California, Riverside

Copyright © 2020 Jauregui et al. This is an open-access article distributed under the terms of the [Creative Commons Attribution 4.0 International license](https://creativecommons.org/licenses/by/4.0/).

Address correspondence to Linda J. Johnson, Linda.Johnson@agresearch.co.nz.

* Present address: Suliana E. Teasdale, Department of Conservation, Wellington, New Zealand.

Received 13 April 2020

Accepted 9 June 2020

Published 2 July 2020

scaffolds with a final genome size of 29.247 Mb, an N_{50} value of 4 Mb, and a 49% GC content. A BUSCO (19) run using the Ascomycota database vodb9 (20) and *Aspergillus nidulans* as the gene predictor template reported a genome completeness of 99.1%, 5 duplicated benchmarking universal single-copy orthologs (BUSCOs) (0.4%), and 5 fragmented BUSCOs (0.4%).

Data availability. This whole-genome shotgun project has been deposited at DDBJ/ENA/GenBank under the accession number [JAALXG000000000](https://doi.org/10.1080/17550874.2019.1610913). The version described in this paper is version JAALXG010000000. The raw Illumina data from BioProject [PRJNA599221](https://doi.org/10.1080/17550874.2019.1610913) were submitted to the NCBI Sequence Read Archive (SRA) under accession numbers [SRX7508659](https://doi.org/10.1080/17550874.2019.1610913) and [SRX7508658](https://doi.org/10.1080/17550874.2019.1610913).

ACKNOWLEDGMENTS

This study was supported by Grasslanz Technology Ltd. and the International Livestock Research Institute (ILRI). The latter was through a contract with the Swedish International Development Cooperation Agency (SIDA) for the program “Innovative Programmatic Approach to Climate Change in Support of Biosciences Eastern and Central Africa’s (BeCA) Mission: Climate-Smart *Brachiaria* Grasses for Improving Livestock Production in East Africa.”

REFERENCES

- Teasdale SE, Caradus JR, Johnson LJ. 2018. Fungal endophyte diversity from tropical forage grass *Brachiaria*. *Plant Ecol Divers* 11:611–624. <https://doi.org/10.1080/17550874.2019.1610913>.
- Nonaka K, Kaifuchi S, Ōmura S, Masuma R. 2013. Five new *Simplicillium* species (Cordycipitaceae) from soils in Tokyo, Japan. *Mycoscience* 54: 42–53. <https://doi.org/10.1016/j.myc.2012.07.002>.
- Zare R, Gams W. 2001. A revision of *Verticillium* section Prostrata. IV. The genera *Lecanicillium* and *Simplicillium* gen. nov. *Nova Hedwigia* 73:1–50.
- Wei D-P, Wanasinghe DN, Hyde KD, Mortimer PE, Xu J, Xiao Y-P, Bhunjun CS, To-Anun C. 2019. The genus *Simplicillium*. *MycKeys* 60:69–92. <https://doi.org/10.3897/mycokeys.60.38040>.
- Chen W-H, Liu C, Han Y-F, Liang J-D, Tian W-Y, Liang Z-Q. 2019. Three novel insect-associated species of *Simplicillium* (Cordycipitaceae, Hypocreales) from Southwest China. *MycKeys* 58:83–102. <https://doi.org/10.3897/mycokeys.58.37176>.
- Ward NA, Schneider RW, Aime MC. 2011. Colonization of soybean rust sori by *Simplicillium lanosoneum*. *Fungal Ecol* 4:303–308. <https://doi.org/10.1016/j.funeco.2011.03.008>.
- Ward NA, Robertson CL, Chanda AK, Schneider RW. 2012. Effects of *Simplicillium lanosoneum* on *Phakopsora pachyrhizi*, the soybean rust pathogen, and its use as a biological control agent. *Phytopathology* 102:749–760. <https://doi.org/10.1094/PHYTO-01-11-0031>.
- Zhao D, Liu B, Li LY, Zhu XF, Wang YY, Wang JQ, Duan YX, Chen LJ. 2013. *Simplicillium chinense*: a biological control agent against plant parasitic nematodes. *Biocontrol Sci Technol* 23:980–986. <https://doi.org/10.1080/09583157.2013.809514>.
- Uchida R, Kondo A, Yagi A, Nonaka K, Masuma R, Kobayashi K, Tomoda H. 2019. Smpotentin, a new potentiator of amphotericin B activity against *Candida albicans*, produced by *Simplicillium minatense* FKI-4981. *J Antibiot (Tokyo)* 72:134–140. <https://doi.org/10.1038/s41429-018-0128-x>.
- Dong Q, Dong R, Xing X, Li Y. 2018. A new antibiotic produced by the cyanobacterium-symbiotic fungus *Simplicillium lanosoneum*. *Nat Prod Res* 32:1348–1352. <https://doi.org/10.1080/14786419.2017.1343320>.
- Fukuda T, Sudoh Y, Tsuchiya Y, Okuda T, Igarashi Y. 2014. Isolation and biosynthesis of preussin B, a pyrrolidine alkaloid from *Simplicillium lanosoneum*. *J Nat Prod* 77:813–817. <https://doi.org/10.1021/np400910r>.
- Liang X, Zhang X-Y, Nong X-H, Wang J, Huang Z-H, Qi S-H. 2016. Eight linear peptides from the deep-sea-derived fungus *Simplicillium obclavatum* EIODSF 020. *Tetrahedron* 72:3092–3097. <https://doi.org/10.1016/j.tet.2016.04.032>.
- Liang X, Nong X-H, Huang Z-H, Qi S-H. 2017. Antifungal and antiviral cyclic peptides from the deep-sea-derived fungus *Simplicillium obclavatum* EIODSF 020. *J Agric Food Chem* 65:5114–5121. <https://doi.org/10.1021/acs.jafc.7b01238>.
- Gomes AAM, Pinho DB, Cardeal ZL, Menezes HC, De Queiroz MV, Pereira OL. 2018. *Simplicillium coffeanum*, a new endophytic species from Brazilian coffee plants, emitting antimicrobial volatiles. *Phytotaxa* 333: 188–298. <https://doi.org/10.11646/phytotaxa.333.2.2>.
- Le Dang Q, Shin TS, Park MS, Choi YH, Choi GJ, Jang KS, Kim IS, Kim J-C. 2014. Antimicrobial activities of novel mannosyl lipids isolated from the biocontrol fungus *Simplicillium lamellicola* BCP against phytopathogenic bacteria. *J Agric Food Chem* 62:3363–3370. <https://doi.org/10.1021/jf500361e>.
- Bolger AM, Lohse M, Usadel B. 2014. Trimmomatic: a flexible trimmer for Illumina sequence data. *Bioinformatics* 30:2114–2120. <https://doi.org/10.1093/bioinformatics/btu170>.
- Hernandez D, François P, Farinelli L, Osterås M, Schrenzel J. 2008. *De novo* bacterial genome sequencing: millions of very short reads assembled on a desktop computer. *Genome Res* 18:802–809. <https://doi.org/10.1101/gr.072033.107>.
- Boetzer M, Henkel CV, Jansen HJ, Butler D, Pirovano W. 2011. Scaffolding pre-assembled contigs using SSPACE. *Bioinformatics* 27:578–579. <https://doi.org/10.1093/bioinformatics/btq683>.
- Simão FA, Waterhouse RM, Ioannidis P, Kriventseva EV, Zdobnov EM. 2015. BUSCO: assessing genome assembly and annotation completeness with single-copy orthologs. *Bioinformatics* 31:3210–3212. <https://doi.org/10.1093/bioinformatics/btv351>.
- Waterhouse RM, Tegenfeldt F, Li J, Zdobnov EM, Kriventseva EV. 2013. OrthoDB: a hierarchical catalog of animal, fungal and bacterial orthologs. *Nucleic Acids Res* 41:358–365. <https://doi.org/10.1093/nar/gks1116>.