

# Draft Genome Sequence of *Alternaria alternata* ATCC 34957

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**We report the draft genome sequence of *Alternaria alternata* ATCC 34957. This strain was previously reported to produce alternariol and alternariol monomethyl ether on weathered grain sorghum. The genome was sequenced with PacBio technology and assembled into 27 scaffolds with a total genome size of 33.5 Mb.**

**Received** 10 November 2015 **Accepted** 18 November 2015 **Published** 14 January 2016

**Citation** Nguyen HDT, Lewis CT, Lévesque CA, Gräfenhan T. 2016. Draft genome sequence of *Alternaria alternata* ATCC 34957. *Genome Announc* 4(1):e01554-15. doi:10.1128/genomeA.01554-15.

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The genus *Alternaria* was originally described based on *A. tenuis* (1). Almost 100 years later, *A. tenuis* and *Torula alternata* (2) were synonymized with *A. alternata* (3). Then, another century later, the genus *Alternaria* was drastically redefined based on multilocus phylogenetic analyses (4). Currently, the species *A. alternata* comprises 35 morphospecies, which cannot be distinguished phylogenetically (5). *A. alternata* has no known sexual stage and produces characteristic dark-colored phaeodictyosporangia in chains with a beak of tapering apical cells. This species is found to infect hundreds of host species of plants, causing leaf spots and other diseases. A wide range of metabolites have been reported for *A. alternata* (6, 7), including alternariol (AOH), alternariol monomethyl ether (AME), altenuene (ALT), altertoxin I, and tenuazonic acid (TEA) in weathered grain sorghum by isolate RL-8442-2 (ATCC 34957) (6, 7). For over 40 years, this isolate has retained its propensity to form these “emerging” mycotoxins in culture (K. Sivagnanam, personal communications). Here, we report the whole-genome sequence of this strain of *A. alternata*, ATCC 34957. This information will be used to discern the synteny of genes involved in the biosynthesis pathways of secondary metabolites.

Genomic DNA was isolated using a large-scale version of a previously described method with an additional 5M/3M potassium acetate/acetic acid step and RNase treatment (8). Extracted DNA was sheared using the HydroShear Plus DNA shearing instrument (Digilab, Marlborough, MA, USA) and then size-selected on a BluePippin system (Sage Science Inc., Beverly, MA, USA). The DNA libraries were prepared following the Pacific Biosciences 20-kb Template Preparation Using BluePippin Size-Selection System protocol. The DNA damage repair, end repair, and SMRT bell ligation steps were performed as described in the template preparation protocol with the SMRTbell template prep kit version 1.0 reagents (Pacific Biosciences, Menlo Park, CA, USA). The libraries were sequenced on a PacBio RSII instrument using the MagBead OneCellPerWell loading protocol, DNA sequencing kit 2.0, SMRT cells version 3, and 3-h movies. *De novo* genome assembly was performed with Celera WGS assembler version 8.2, using the PBcR pipeline for high-noise single-molecule sequencing data (9). Genome statistics were determined with

QUAST version 2.3 (10). CEGMA version 2.5 (11) was run on the scaffolds to detect the percentage of conserved eukaryotic genes.

*A. alternata* has 9 to 11 chromosomes and a genome of up to 33.6 Mb (12). Our *A. alternata* ATCC 34957 genome is contained in 27 scaffolds spanning 33.5 Mb with 16× coverage. The largest scaffold is 3.97 Mb and the  $N_{50}$  is 2.83 Mb. Scores of 96% and 97% were obtained from the complete and partial gene sets, respectively, using CEGMA. This is a draft genome but close to being complete because the telomeric sequence motif (13, 14) was found at both ends of four scaffolds (scaffolds 8, 9, 10, and 11) and at one end of seven scaffolds (scaffolds 2, 3, 4, 5, 7, 12, and 13).

**Nucleotide sequence accession numbers.** This whole-genome shotgun project has been deposited at DDBJ/EMBL/GenBank under accession number [LMXP000000000](https://www.ncbi.nlm.nih.gov/nuccore/LMXP000000000). The version described in this paper is the first version, LMXP010000000.

## ACKNOWLEDGMENTS

We thank Geneviève Geneau and Patrick Willett at the McGill University and Genome Québec Innovation Centre for the fine sequencing service provided for this study. The assistance of Janice Bamforth, Susan Patrick, and Tigst Demeke is also greatly appreciated. We declare no conflict of interests.

Genome sequencing of ATCC 34957 was funded by Growing Forward 2 from the Canadian Grain Commission and Agriculture & Agri-Food Canada, with additional support from Canadian Safety and Security Program grant CRTI 09-462RD/CSSP 30vv01. The funders had no role in study design, data collection, and interpretation, or the decision to submit the work for publication.

## FUNDING INFORMATION

Growing Forward 2 provided funding to Hai D. T. Nguyen and Tom Gräfenhan. Canadian Safety and Security Program provided funding to Christopher T. Lewis, André Lévesque, Tom Gräfenhan under grant number CSSP 30vv01/CRTI 09-462RD.

## REFERENCES

1. Nees von Esenbeck CGD. 1816. Das System der Pilze und Schwämme. Stahel, Wurzburg, Germany.
2. Fries EM. 1832. Systema mycologicum, vol 3. E. Moritz, Greifswald, Germany.

3. Von Keissler K. 1912. Zur kenntnis der pilzflora krains. Beihefte zum Botanischen Zentralblatt 29:395–440.
4. Woudenberg JHC, Groenewald JZ, Binder M, Crous PW. 2013. *Alternaria* redefined. Stud Mycol 75:171–212. <http://dx.doi.org/10.3114/sim0015>.
5. Woudenberg JHC, Seidl MF, Groenewald JZ, de Vries M, Stielow JB, Thomma BPHJ, Crous PW. 2015. *Alternaria* section *alternaria*: species, *formae speciales* or pathotypes? Stud Mycol 82:1–21. <http://dx.doi.org/10.1016/j.simyco.2015.07.001>.
6. Burroughs R, Seitz LM, Sauer DB, Mohr HE. 1976. Effect of substrate on metabolite production of *alternaria alternata*. Appl Environ Microbiol 31:685–690.
7. Seitz LM, Sauer DB, Mohr HE, Burroughs R. 1975. Weathered grain sorghum: natural occurrence of alternariols and storability of the grain. Phytopathology 65:1259–1263. <http://dx.doi.org/10.1094/Phyto-65-1259>.
8. McDonald MB, Elliot LJ, Sweeney PM. 1994. DNA extraction from dry seeds for RAPD analyses in varietal identification studies. Seed Sci Technol 22:171–176.
9. Berlin K, Koren S, Chin C, Drake JP, Landolin JM, Phillippy AM. 2015. Assembling large genomes with single-molecule sequencing and locality-sensitive hashing. Nat Biotechnol 33:623–630. <http://dx.doi.org/10.1038/nbt.3238>.
10. Gurevich A, Saveliev V, Vyahhi N, Tesler G. 2013. QAST: quality assessment tool for genome assemblies. Bioinformatics 29:1072–1075. <http://dx.doi.org/10.1093/bioinformatics/btt086>.
11. Parra G, Bradnam K, Korf I. 2007. CEGMA: a pipeline to accurately annotate core genes in eukaryotic genomes. Bioinformatics 23:1061–1067. <http://dx.doi.org/10.1093/bioinformatics/btm071>.
12. Akamatsu H, Taga M, Kodama M, Johnson R, Otani H, Kohmoto K. 1999. Molecular karyotypes for *alternaria* plant pathogens known to produce host-specific toxins. Curr Genet 35:647–656. <http://dx.doi.org/10.1007/s002940050464>.
13. Lefebvre F, Joly DL, Labbe C, Teichmann B, Linning R, Belzile F, Bakkeren G, Belanger RR. 2013. The transition from a phytopathogenic smut ancestor to an anamorphic biocontrol agent deciphered by comparative whole-genome analysis. Plant Cell 25:1946–1959. <http://dx.doi.org/10.1105/tpc.113.113969>.
14. Wu C, Kim Y-S, Smith KM, Li W, Hood HM, Staben C, Selker EU, Sachs MS, Farman ML. 2009. Characterization of chromosome ends in the filamentous fungus *Neurospora crassa*. Genetics 181:1129–1145. <http://dx.doi.org/10.1534/genetics.107.084392>.