Draft Genome Resources for the Phytopathogenic Fungi *Monilinia fructicola*, *M. fructigena*, *M. polystroma*, and *M. laxa*, the Causal Agents of Brown Rot

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Abstract

Fungi in the genus *Monilinia* cause brown rot disease of stone and pome fruits. Here, we report the draft genome assemblies of four important phytopathogenic species: *M. fructicola*, *M. fructigena*, *M. polystroma*, and *M. laxa*. The draft genome assemblies were 39 Mb (*M. fructigena*), 42 Mb (*M. laxa*), 43 Mb (*M. fructicola*), and 45 Mb (*M. polystroma*) with as few as 550 contigs (*M. laxa*). These are the first draft genome resources publicly available for *M. laxa*, *M. fructigena*, and *M. polystroma*.

Monilinia fructicola, *M. fructigena*, *M. polystroma*, and *M. laxa* cause brown rot of stone (e.g., peach, nectarine, cherry, and plum) and some pome (apple and pear) fruits. These fungi can cause twig and blossom blights, as well as economically important fruit rot as the fruit ripen and their sugar content increases. *M. fructicola* and *M. laxa* are established within the United States, but *M. fructigena* and *M. polystroma* are currently not known from this country. Diagnostic methods for phytosanitary purposes need to be quick and reliable. Morphological similarity among these species and their overlapping host ranges can make differentiation between native and exotic species problematic and time-consuming. Whole-genome sequences of these four important pathogenic species will allow researchers to perform comparative genomic analysis to aid in the development of diagnostic markers to discriminate among them. In this study, we performed whole-genome sequencing and de novo assemblies of these four *Monilinia* species of interest.

We cultured all *Monilinia* isolates (Table 1) in potato dextrose broth for 4 to 6 days at 22 ± 2°C on a rotary shaker at 120 rpm and harvested mycelia by filtration. We cryogenically pulverized mycelia and isolated genomic DNA using a CTAB method modified from that in Porebski et al. (1997). PCR amplification of the internal transcribed spacer (ITS) and BLAST comparison confirmed the species identity for all four fungal cultures. Genomic DNA libraries were constructed using the TruSeq Nano Library Prep Kit (NeoPrep for *M. fructigena* and *M. laxa*) and per the protocol provided by Illumina for *M. fructicola* and *M. polystroma*. All libraries were quantified with a Qubit version 3.0 fluorometer (Life Technologies). Library quality was evaluated using the Agilent Genomic ScreenTape Assay (Agilent Technologies). All

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Table 1. Isolate information and draft genome assembly summary statistics for *Monilinia* spp.

Species	Isolate name	Origin	Year	Provided by	Read count	Number of contigs	Maximum scaffold length (bp)	N50	Genome coverage	GC content (%)	Total length (bp)
<i>M. fruticola</i>	BR-32	Peach, Berrien Co., MI	2009	Sundin	42,408,212	2,350	414,517	66,420	296×	41.92	42,827,339
<i>M. fructigena</i>	Mfg-5-SP-A	Apple, Hungary	2009	Schnabel	20,423,570	1,635	260,257	56,695	157×	41.90	39,335,235
<i>M. laxa</i>	EBR Ba11b	Cherry, Niagara, NY	2008	Schnabel	6,455,926	550	673,002	159,077	46×	40.28	41,849,271
<i>M. polystroma</i>	SP-61	Plum, Skierniewice, Poland	2010	Poniatowska	34,783,998	1,189	621,141	140,651	232×	39.27	44,639,555

libraries were sequenced on a MiSeq (Illumina) using a MiSeq version 3.0 reagent kit (300-bp × 2). Paired-end reads were checked for quality using FASTQC (Andrews 2016) before and after trimming adapters in CLC Workbench (CLC Genomics Workbench 10.1.1 <https://www.qia-genbioinformatics.com/>). Reads were de novo assembled using CLC Workbench. The total number of reads ranged from 6 to 42 million and were assembled in as little as 550-contigs for the *M. laxa* genome and up to 2,350-contigs for the *M. fruticola* genome (Table 1). Genome assembly completeness using the benchmarking universal single-copy orthologs (BUSCO) was estimated to be 97 to 98% for all four species (<http://busco.ezlab.org>) (Simão et al. 2015). Genome coverage for *M. fruticola*, *M. fructigena*, and *M. polystroma* was over 150× while only 46× for *M. laxa*. Draft genome assemblies are estimated between 39 Mb (*M. fructigena*) to 45 Mb (*M. polystroma*). These genome assemblies are of comparable size to other published genomes in the family Sclerotiniaceae (e.g., *Sclerotinia sclerotiorum* 38.8 Mb [Derbyshire et al. 2017] and *Botrytis cinerea* 42.6 Mb [Van Kan et al. 2017], and most recently a different strain of *M. fruticola* 44.6 Mb [NGKE01000001] and *M. aucupariae* 54.9 Mb [NGKF01000001]).

The genomic resources reported here expand our molecular knowledge of *Monilinia* spp. and make it possible to conduct comparative genomics studies to identify species-specific gene targets and genetic variants for better diagnostic tools.

Accession numbers. This whole-genome shotgun project has been deposited at GenBank under the accession numbers PDME000000000 (*M. fruticola*), PDMD000000000 (*M. fructigena*), PDMC000000000 (*M. polystroma*), and PDUE000000000 (*M. laxa*). The raw data for this experiment have been submitted to NCBI with SRA study SRP121347. Voucher specimens have been submitted to the U.S. National Fungus Collections under BPI910582 (*M. polystroma*), BPI910583 (*M. fructigena*), BPI910584 (*M. laxa*), and BPI910585 (*M. fruticola*).

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