Genome Sequences of Three Races of *Peronospora effusa*: A Resource for Studying the Evolution of the Spinach Downy Mildew Pathogen

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Abstract

Downy mildew disease, caused by the obligate oomycete pathogen *Peronospora effusa*, is the most important economic constraint for spinach production. Three races (races 12, 13, and 14) of *P. effusa* have been sequenced and assembled. The draft genomes of these three races have been deposited to GenBank and provide useful resources for dissecting the interaction between the host and the pathogen and may provide a framework for determining the mechanism by which new races of the pathogen are rapidly emerging.

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Genome Announcement

Spinach is a popular and nutritious vegetable (Morelock and Correll 2008). Current high-density plant populations for fresh-market spinach production provide conditions highly conducive for disease development. Downy mildew disease, caused by the obligate pathogen *Peronospora effusa* (= *P. farinosa* f. sp. *spinaciae*), is the most economically important disease of spinach (Correll et al. 2011). The use of resistant cultivars to manage downy mildew applies strong selection pressure on the pathogen, and 14 of the 17 races of *P. effusa* have been identified in recent years (Feng et al. 2014, 2018; Irish et al. 2003, 2007). The rapid appearance of new races of this pathogen make it a challenge to use resistance as a sustainable management tool, threatening spinach production. However, the mechanism of virulence evolution in *P. effusa* is largely unknown. Sequencing the genome of this pathogen will ultimately improve the fundamental understanding of the genes involved in virulence and the mechanisms underlying host–pathogen interactions and pathogen evolution.

P. effusa is an obligate pathogen and cannot be grown in axenic culture. Thus, sporangia of three strains of *P. effusa* (UA2209, UA0510C, and UA4410, representing races 12, 13, and 14, respectively) (Feng et al. 2014) were recovered from infected spinach leaves by washing sporangia off with deionized water and concentrated by centrifugation before DNA extraction. Total genomic DNA of each strain was isolated using the cetyltrimethylammonium bromide method. After enzymatic fragmentation, 200-bp libraries were constructed and then ran in one lane of 100-bp paired-end HiSeq2000. Strain UA0510C was sequenced at the Genomics Core Facility, Purdue University (West Lafayette, IN, U.S.A.), and UA2209 and UA4410 were sequenced at BGI (Shenzhen, China). Approximately 128 to 148 million 100-bp paired-end reads (31 to 35 gigabases) were obtained for each strain (Table 1). In order to remove any nontarget sequences from other sources such as epiphytic bacteria, the raw reads were mapped to a set of 483-Mb oomycete reference genome sequences (downloaded from GenBank on 8 October 2015) with

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Table 1. Assembly statistics of the three races of *Peronospora effusa*

Assembly	Race 12 (UA2209)	Race 13 (UA0510C)	Race 14 (UA4410)
Raw read (Mb)	33,456	31,668	34,682
Percentage of reads that match oomycete	84.35	61.68	81.43
Number of contigs	4,060	4,384	4,020
Largest contig (bp)	141,591	168,434	131,290
Total length (bp)	25,220,668	23,993,179	24,880,370
GC (%)	46.9	46.88	46.91
N50	21,343	18,134	22,141
L50	340	376	327

Burrows-Wheeler Aligner (BWA) software using a short-read algorithm (Li and Durbin 2009); then, the mapped raw reads, approximately 62 to 84% of the original total, were assembled using CLC Genomics Workbench 7 with de Bruijn graph assembly. The draft genome of each strain was approximately 25 Mb, which represented approximately 50% of the estimated genome size but approximately 78% of the estimated genome size of a race 1 strain previously reported (Klein et al. 2017). When the completeness of the genome sequences was assessed by comparing the draft genome sequences to the 290 eukaryotic single-copy orthologs using the software BUSCO (Waterhouse et al. 2018), the results indicated that 82, 81, and 83% of the genomes were complete or partially complete (71, 68, and 72% were complete and 11, 13, and 11% were partially finished) for races 12, 13, and 14 of *P. effusa*, respectively. The incompleteness may be due to loss of *P. effusa*-specific reads when only reads mapped to the oomycete reference genome were used for assembly. The draft genomes comprised approximately 4,000 to 4,400 contigs with the GC content of 46.9%. More than 50% of the sequences were contributed by 327 to 376 contigs (L50) that were 18 kb or longer (N50), with the largest contigs being 141, 168, and 131 kb for strains UA2209, UA0510C, and UA4410, respectively (Table 1). More than 15,000 single-nucleotide polymorphisms (SNP) were identified from the three sequenced genomes using BWA and SAMtools (Li et al. 2009). Forty-six selected SNP have been used to investigate the population diversity of *P. effusa* and comparative genomic analysis revealed that both sexual and asexual reproduction may have generated novel genotypes with unique virulence phenotypes. (Lyon et al. 2016).

This whole-genome shotgun project has been deposited in DDBJ/ENA/GenBank under the accession numbers NPIT000000000, NPIU000000000, and NPIV000000000. The versions described in this article are the first versions (NPIT01000000, NPIU01000000, and NPIV01000000).

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Author-Recommended Internet Resource

Genomics Workbench 7: <https://www.qiagenbioinformatics.com/>