



Draft Genome Sequences of 12 Isolates from 3 *Fusarium* Species Recovered from Moldy Peanuts

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ABSTRACT In this report, we announce the sequences of six genomes of *Fusarium proliferatum* (isolates MOD1-FUNGI8, -12, -13, -14, -15, and -19), four genomes of *Fusarium oxysporum* (MOD1-FUNGI9, -10, -11, and -16), and two genomes of the *Fusarium incarnatum-Fusarium equiseti* species complex (MOD1-FUNGI17 and MOD1-FUNGI18) isolated from moldy peanuts from the Washington, DC, area.

Foods of plant origin, such as tree nuts, are known to foster the growth of various microorganisms, including toxigenic and pathogenic fungi (1). Members of the genera *Aspergillus*, *Penicillium*, and *Fusarium* are known to be the major mycotoxin-producing fungi (2–4). According to past research, peanuts and tree nuts, such as walnuts, pistachios, and pecans, are frequently colonized by these molds (1). *Fusarium* species are among the most prevalent toxin-producing fungi and plant pathogens, causing crown rot, head blight, and scab on cereal grains and vascular wilts on a wide range of horticultural crops (5–10). In addition, fusaria produce diverse mycotoxins, including trichothecenes (T-2 toxin, HT-2 toxin, deoxynivalenol, and nivalenol), zearalene, and fumonisins, and other toxic secondary metabolites (3, 4). These toxins pose a threat to food safety and human health if they enter the food chain. Therefore, comparative genomics provides a means to accurately catalog their pathogenic potential.

In-shell peanuts were tested for the presence of live fungi by direct plating onto dichloran glycerol (DG18) agar as described in the Bacteriological Analytical Manual (BAM), Chapter 18 (11). Colonies were randomly selected (usually one or two of each morphological type were picked). The isolates were microscopically examined and identified to the genus level using conventional culture methods and taxonomical keys (12). The recovered molds were purified by subculturing on potato dextrose agar (BD Difco, Detroit, MI) and incubated at 25°C for 5 days. Mycelium for DNA extraction was obtained by culturing each strain in potato dextrose broth at 25°C for 48 h. Subsequently, the DNA was extracted with the AllPrep fungal DNA/RNA/protein kit (Qiagen, Germantown, MD), following the manufacturer's instructions. The quality and purity of the genomic DNA (gDNA) was assessed spectrophotometrically using a Nanodrop spectrophotometer (Thermo Scientific, Wilmington, DE), and quality control was performed using a Qubit 2.0 fluorometer (Life Technologies, Burlington, Canada). Whole-genome sequencing was performed using a Nextera XT DNA library prep kit (Illumina, Inc., San Diego, CA) with 2 × 150-bp paired-end sequencing on an Illumina NextSeq sequencer with a NextSeq 500/550 midoutput reagent cartridge V2 ($n = 8$). Fast QC (Q score, >30) was used to check the raw sequence data for quality control, followed by *de novo* assembly using SPAdes 3.8.2 (Center for Algorithmic Biotechnology, St. Petersburg State University, St. Petersburg, Russia) (13). The draft genomes comprised between 325 and 2,370 contigs, with an N_{50} value that ranged from 63,924 to 262,090 bp,

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TABLE 1 *Fusarium* species statistics data

BioSample accession no.	Isolate	Species	GenBank accession no.	SRA accession no.	No. of contigs	No. of reads	Genome size (bp)	N ₅₀ value	Avg coverage (x)	G+C content (%)
SAMN10078280	MOD1-FUNG18	<i>F. proliferatum</i>	RBJG000000000	SRR7889975	1,159	33,254,726	43,155,960	68,427	27	48.8
SAMN10078281	MOD1-FUNG19	<i>F. oxysporum</i>	RBCG000000000	SRR7889959	2,370	37,245,070	47,450,651	63,924	28	48.3
SAMN10078282	MOD1-FUNG10	<i>F. oxysporum</i>	RBJF000000000	SRR7889971	1,950	37,008,864	46,252,701	66,395	28	48.4
SAMN10078283	MOD1-FUNG11	<i>F. oxysporum</i>	RBCF000000000	SRR7889970	1,377	55,555,690	46,282,089	176,916	82	48.4
SAMN10078284	MOD1-FUNG12	<i>F. proliferatum</i>	RBCF000000000	SRR7889969	562	50,008,512	43,124,502	172,812	79	48.8
SAMN10078285	MOD1-FUNG13	<i>F. proliferatum</i>	RBCD000000000	SRR7889968	1,006	33,134,216	43,139,311	84,221	40	48.8
SAMN10078286	MOD1-FUNG14	<i>F. proliferatum</i>	RBCD000000000	SRR7889967	563	28,820,800	43,147,616	181,851	44	48.8
SAMN10078287	MOD1-FUNG15	<i>F. proliferatum</i>	RBCB000000000	SRR7889966	578	71,676,280	43,337,992	253,187	106	48.7
SAMN10078288	MOD1-FUNG16	<i>F. oxysporum</i>	RBCA000000000	SRR7889965	944	73,866,756	44,448,977	262,090	110	48.3
SAMN10078289	MOD1-FUNG17	<i>Fusarium</i> sp. FIESC_29	RBJE000000000	SRR7889964	325	37,761,252	38,264,798	228,787	23	48.5
SAMN10078290	MOD1-FUNG18	<i>Fusarium</i> sp. FIESC_29	RBBZ000000000	SRR7889963	340	46,941,284	38,253,301	220,485	26	48.5
SAMN10078291	MOD1-FUNG19	<i>F. proliferatum</i>	RBBY000000000	SRR7889962	449	54,664,676	43,132,928	195,358	50	48.8

and the depth of coverage ranged from 23× to 110× (Table 1). The genome sizes for *Fusarium proliferatum* ranged from 43,155,960 to 43,337,992 bp, for *Fusarium oxysporum* from 44,448,977 to 47,450,651 bp, and for *Fusarium incarnatum-Fusarium equiseti* species complex (FIESC 29) from 38,253,301 to 38,264,798 bp (Table 1). A custom kmer analysis of the reads determined an initial identification of the species that was confirmed by matching the TEF1 gene sequences from the assemblies against the *Fusarium* multilocus sequence type (MLST) database (14).

Data availability. The draft genome assemblies were deposited in DDBJ/ENA/GenBank under BioProject number [PRJNA482816](https://www.ncbi.nlm.nih.gov/bioproject/PRJNA482816), and the whole-genome sequencing (WGS) and SRA accession numbers for the genomes are listed in the Table 1.

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REFERENCES

1. Tournas VH, Niaz NS, Kohn JS. 2015. Fungal presence in selected tree nuts and dried fruits. *Microbiol Insights* 8:1–6. <https://doi.org/10.4137/MBI.S24308>.
2. Villani A, Moretti A, De Saeger S, Han Z, Di Mavungu JD, Soares CMG, Proctor RH, Venâncio A, Lima N, Stea G, Paciolla C, Logrieco AF, Susca A. 2016. A polyphasic approach for characterization of a collection of cereal isolates of the *Fusarium incarnatum-equiseti* species complex. *Int J Food Microbiol* 234:24–35. <https://doi.org/10.1016/j.jfoodmicro.2016.06.023>.
3. Wagacha JM, Muthomi JW. 2008. Mycotoxin problem in Africa: current status, implications to food safety and health and possible management strategies. *Int J Food Microbiol* 124:1–12. <https://doi.org/10.1016/j.jfoodmicro.2008.01.008>.
4. Darwish WS, Ikenaka Y, Nakayama SM, Ishizuka M. 2014. An overview on mycotoxin contamination of foods in Africa. *J Vet Med Sci* 76:789–797. <https://doi.org/10.1292/jvms.13-0563>.
5. Booth C. 1971. The genus *Fusarium*. Commonwealth Mycology Institute, Kew, Surrey, England.
6. Latiffah Z, Nur H, Baharuddin S, Maziah Z. 2009. Morphological and RAPD analysis of *Fusarium* species associated with root and stem rot of *Dendrobium* orchid in Northern Peninsula Malaysia. *HAYATI J Biosci* 16:64–68.
7. Chang M, Hyun IK, Lee YH, Lee DH. 1998. Leaf spot of *Cymbidium hybridum* caused by *Fusarium proliferatum*. *Korean J Plant Pathol* 14:664–667.
8. Ichikawa K, Aoki T. 2000. New leaf spot disease of *Cymbidium* species caused by *Fusarium subglutinans* and *Fusarium proliferatum*. *J Gen Plant Pathol* 66:213–218. <https://doi.org/10.1007/PL00012948>.
9. Kim WG, Lee BD, Cho WD, Sung JM. 2002. Occurrence of dry rot on *Cymbidium* orchids caused by *Fusarium* spp. in Korea. *Korean J Plant Pathol* 18:156–160. <https://doi.org/10.5423/PPJ.2002.18.3.156>.
10. Nelson PE, Dignani MC, Anaissie EJ. 1994. Taxonomy, biology, and clinical aspects of *Fusarium* species. *Clin Microbiol Rev* 7:479–504. <https://doi.org/10.1128/CMR.7.4.479>.
11. Tournas V, Stack ME, Mislivec PB, Koch HA, Bandler R. 2001. Chapter 18, Yeasts, molds and mycotoxins. In *Bacteriological analytical manual (BAM)*. Food and Drug Administration, Silver Spring, MD. <https://www.fda.gov/food/foodscienceresearch/laboratorymethods/ucm2006949.htm>.
12. Nelson PE, Toussoun TA, Marasas WFO. 1983. *Fusarium* species: an illustrated manual for identification. The Pennsylvania State University Press, University Park, Pennsylvania.
13. Bankevich A, Nurk S, Antipov D, Gurevich AA, Dvorkin M, Kulikov AS, Lesin VM, Nikolenko SI, Pham S, Prjibelski AD, Pyshkin AV, Sirotkin AV, Vyahhi N, Tesler G, Alekseyev MA, Pevzner PA. 2012. SPAdes: a new genome assembly algorithm and its applications to single-cell sequencing. *J Comput Biol* 19:455–477. <https://doi.org/10.1089/cmb.2012.0021>.
14. Geiser DM, Jiménez-Gasco M, Kang S, Makalowska I, Veeraraghavan N, Ward TJ, Zhang N, Kuldau GA, O'Donnell K. 2004. FUSARIUM-ID v. 1.0: a DNA sequence database for identifying *Fusarium*. *Eur J Plant Pathol* 110:473–479. <https://doi.org/10.1023/B:EJPP.0000032386.75915.a0>.