



Draft Genome Sequences of 20 *Aspergillus flavus* Isolates from Corn Kernels and Cornfield Soils in Louisiana

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ABSTRACT *Aspergillus flavus* is a common saprophyte and opportunistic fungal pathogen that infects plants, animals, and humans. It also produces numerous toxic and nontoxic secondary metabolites. Here, we report the draft genome sequences of 20 *A. flavus* isolates, belonging to 16 vegetative compatibility groups, from Louisiana corn kernels and cornfield soils.

Aspergillus *flavus* is an opportunistic pathogen that infects plants, animals, and humans and produces aflatoxin, a natural carcinogen (1–4). Aflatoxin consumed in grains or milk is acutely toxic, stunts children's growth, and induces liver cancer (1, 5–8). *Aspergillus flavus* has the potential to produce several toxic secondary metabolites beyond aflatoxins B₁ and B₂, including cyclopiazonic acid, kojic acid, β-nitropropionic acid, aspertoxin, aflatrem, and aspergillic acid (1–4). The fungus may also cause allergic reactions in humans and aspergillosis diseases, notably in immunocompromised people's lungs (5–8). Under favorable hot and dry field conditions, *A. flavus* can cause corn ear rot, which is frequently associated with aflatoxin contamination inside diseased corn kernels (9–12). Aflatoxin contamination in corn threatens consumer food safety and grower economic stability, causing an estimated \$52 million to over \$1 billion of economic loss per year in the United States (12). In Louisiana, a severe corn aflatoxin outbreak in 1998 resulted in almost total crop loss, and periodic aflatoxin outbreaks still occur (<https://www.farmprogress.com/aflatoxin-levels-low-louisiana-corn>). *Aspergillus flavus* populations are very diverse and may consist of many different vegetative compatibility groups (VCGs) (2, 3, 9, 13–15). Members within a VCG share identical *het* locus alleles and belong to quasiconalineage lineages exhibiting similar quantities of aflatoxin production (some VCGs do not produce any aflatoxins), sclerotial sizes, and mating types (2, 3, 9, 13, 15). *Aspergillus flavus* was isolated on differential/selective medium from 50 soil samples (255 isolates) and 25 surface-sterilized corn kernels (612 isolates) from 85 individual ears collected at maturity from 11 cornfields in Louisiana (15). The isolates belonged to at least 16 VCGs (15). A selection of 20 isolates encompassing all 16 VCGs were deposited in the USDA-SRRC's culture collection.

To prepare for sequencing, 75-ml potato dextrose broth conidial cultures were shaken overnight at 125 rpm and 30°C. Mycelial pellets were collected, flash frozen with liquid nitrogen, and freeze-dried, and DNA was extracted with a Quick-DNA fungal/bacterial DNA miniprep kit (Zymo Research, Irvine, CA). Genomic DNA (gDNA) quality and purity were assessed using a NanoDrop spectrophotometer (Thermo Scientific, Wilmington, DE), and quality control was performed using a Qubit 2.0 fluorometer (Life Technologies, Burlington, Canada). Sequencing libraries for each gDNA extract were prepared using a Nextera XT DNA sample prep kit (Illumina, San Diego, CA) in 2 × 150-bp paired-end format and pooled in a single NextSeq 500/550 midoutput kit v2 ($n = 8$) for sequencing with an Illumina NextSeq sequencer. Low-quality reads were trimmed to a quality threshold of $Q > 30$ using Trimmomatic (16) with a NexteraPE

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TABLE 1 *Aspergillus flavus* strains, aflatoxin production, and genomic statistics data

Genome identifier	Strain/ VCG	Aflatoxigenicity	Aflatoxin B ₁ production (ppb) ± SE ^b	No. of contigs	No. of reads	Genome size (bp)	N ₅₀ (bp)	Coverage (×)	% G+C	SRA accession no.	GenBank accession no.
MOD1-573	RRS4	Aflatoxigenic	11,900 ± 836	670	34,952,640	37,023,989	132,464	42	48.3	SRR11596619	JABVYR0000000000
MOD1-575	RRS7	Aflatoxigenic	20 ± 6	624	31,401,164	36,757,764	142,633	39	48.3	SRR11596618	JABVYS0000000000
MOD1-576	RRS5	Aflatoxigenic	24,006 ± 3,918	688	34,833,608	37,068,150	126,389	42	48.2	SRR11596607	JABVYT0000000000
MOD1-578	RRS9	Aflatoxigenic	3,872 ± 1,026	652	40,610,742	37,193,948	152,925	48	48.3	SRR11596606	JABVYU0000000000
MOD1-580	RRS1	Aflatoxigenic	< 0	683	35,257,442	36,938,434	121,985	42	48.3	SRR11596605	JABVYY0000000000
MOD1-581	RRS10	Nonaflatoxigenic	0 ± 0	723	39,417,907	36,954,782	112,933	39	48.3	SRR11596604	JABVYW0000000000
MOD1-584	RRS11	Aflatoxigenic	1,714 ± 120	595	29,738,152	36,865,287	144,290	41	48.3	SRR11596603	JABVYX0000000000
MOD1-586	RRS3	Aflatoxigenic	16,162 ± 150	756	35,670,908	37,089,482	143,643	42	48.3	SRR11596602	JABVYY0000000000
MOD1-587	RRS12	Aflatoxigenic	42,548 ± 9,686	701	30,328,260	36,985,390	136,341	40	48.3	SRR11596601	JABVYZ0000000000
MOD1-590	RRS2	Aflatoxigenic	14,032 ± 4,858	635	32,785,962	36,946,864	138,751	45	48.3	SRR11596600	JABVZA0000000000
MOD1-591	RRS13	Nonaflatoxigenic	0 ± 0	626	35,283,328	36,626,824	141,548	49	48.3	SRR11596617	JABVZB0000000000
MOD1-595	RRS14	Nonaflatoxigenic	0 ± 0	623	32,152,852	36,915,067	152,851	45	48.2	SRR11596616	JABVZC0000000000
MOD1-599	RRS8	Aflatoxigenic	27,998 ± 9,260	723	38,755,062	37,006,081	152,745	42	48.2	SRR11596615	JABVZD0000000000
MOD1-601	RRS5	Aflatoxigenic	19,114 ± 332	867	40,051,400	37,973,590	170,945	57	48.2	SRR11596614	JABVZE0000000000
MOD1-605	RRS6	Nonaflatoxigenic	0 ± 0	627	36,745,078	36,775,823	155,768	52	48.2	SRR11596613	JABVZF0000000000
MOD1-607	RRS15	Aflatoxigenic	3,968 ± 432	1,167	21,467,712	38,231,283	85,770	30	48.2	SRR11596612	JABVZG0000000000
MOD1-618	RRSSOLO ^a	Aflatoxigenic	12,460 ± 2,650	546	36,203,968	36,963,370	169,107	51	48.3	SRR11596611	JABVZH0000000000
MOD1-619	RRS1	Aflatoxigenic	35	908	39,434,866	36,935,936	129,357	110	48.3	SRR11596610	JABVZI0000000000
MOD1-620	RRS1	Aflatoxigenic	3	1,226	42,922,302	37,016,679	143,589	108	48.2	SRR11596609	JABVZJ0000000000
MOD1-621	RRS1	Aflatoxigenic	3	1,660	37,110,996	36,895,533	138,343	105	48.3	SRR11596608	JABVZK0000000000

^a RRSSOLO refers to an isolate that did not complement any others and therefore was in a singleton VCG.

^b Aflatoxin B₁ (AFB₁) was quantified with ultraperformance liquid chromatography from each of three independent 4-day-old cultures grown in glucose-salts medium. AFB₂ was detected in lower quantities for each extract with AFB₁. MOD1-619, MOD1-620, and MOD1-621 were previously quantified from single cultures grown on rice (15).

adapter file. Trimmed reads were subjected to *de novo* assembly using the SPAdes assembler v3.12.0 (17), and assembly quality assessment utilized QUAST (18). Default settings were used for all software programs unless otherwise noted.

Data availability. Genome sequences of these 20 *Aspergillus flavus* isolates were deposited in DDBJ/ENA/GenBank under BioProject number PRJNA482816. The genome sequencing and assembly statistics (contig number, read number, genome size, N₅₀ value, G+C content, and SRA and whole-genome sequence [WGS] accession numbers) for each isolate are shown in Table 1. This announcement reports the first versions of these genome sequences.

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The views expressed in this article are those of the authors and do not necessarily reflect the official policy of the Department of Health and Human Services, the FDA, or the U.S. Government. References to commercial materials, equipment, or processes do not in any way constitute endorsement.

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