


Complete mitochondrial genome sequence of *Aspergillus flavus* SRRC1009: insight of intraspecific variations on *A. flavus* mitochondrial genomes

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ABSTRACT

The mitogenome of *Aspergillus flavus* SRRC1009 was sequenced to investigate intraspecific variations on mitochondrial genomes of *A. flavus*. It shows 29,202 bp with a typical configuration of *Aspergillus* mitogenome. Sixteen SNPs and 22 INDELS and 17 SNPs and 27 INDELS were identified against AflaGuard[®] and JQ355000, respectively. Phylogenetic trees present in the three *A. flavus* mitochondrial genomes were clustered with *A. oryzae* mitochondrial genome in one clade.

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Aspergillus flavus produces aflatoxin, a notorious mycotoxin that can cause liver cancer (Squire 1981). To date many genome sequences of *A. flavus* were sequenced (Payne et al. 2006; Nierman et al. 2015; Weaver et al. 2017); however, only three mitogenomes are available (Joardar et al. 2012; Yan et al. 2016; Park et al., [under review](#)). To understand intraspecific variations on *A. flavus* mitogenomes, we completed the mitogenome of *A. flavus* SRRC1009.

DNA was extracted using the optimized protocol (Lee et al. 2017). Raw data generated by HiSeq2500 and *de novo* assembly were conducted by Velvet v1.2.10 (Zerbino and Birney 2008). Gap filling was done by SOAPGapCloser v1.12 (Zhao et al. 2011). Basepair confirmation was done by BWA v0.7.17 and SAMtools v1.9 (Li et al. 2009; Li 2013) under the environment of Genome Information System (GeIS; <http://geis.infoboss.co.kr/>). Geneious R11 11.0.5 (Biomatters Ltd, Auckland, New Zealand) was used to annotate its mitogenome with *A. flavus* (MT335777; Park et al., [under review](#)). Voucher sample isolated from barley in Denmark was deposited into South Regional Research Center of USDA (SRRC1009; Klich and Pitt 1988).

Aspergillus flavus SRRC1009 mitogenome length (GenBank accession is MT354769) is 29,202 bp, the shortest mitogenomes among three *A. flavus* mitogenomes. Numbers of PCGs, tRNAs, and rRNAs are 17, 27, and 2, respectively. GC ratio is 26.2%, similar to the two *A. flavus* mitogenomes.

The first mitogenome of *A. flavus* (Yan et al. 2016) presented different features in various aspects including length

(31,602 bp) and GC ratio (25.2%). Based on our mitogenome, this might be another *Aspergillus* species together with the recent study (Park et al., [under review](#)) and phylogenetic tree (Figure 1(A)).

Sixteen SNPs and 22 INDELS were identified in comparison to AflaGuard[®] mitogenome (MT335777; Figure 1(B)). Five non-synonymous SNPs (nsSNPs) were identified in three genes, *NAD1* (two nsSNPs), the hypothetical protein (two nsSNPs), and *NAD3* (one nsSNP). Four synonymous SNPs (sSNPs) were identified in *COX3* (one sSNP), *NAD5* (one sSNP), and *COX1* (two sSNPs). In addition, 17 SNPs and 27 INDELS were found against JQ355000, one more sSNP was found in *NAD5* and number of SNPs in genes is same to those of the previous comparison (Figure 1(B)). Number of variations found in this study occupied small proportion on mitogenomes in comparison to the other fungal species, including *Fusarium oxysporum* (Kwon et al. 2019) as well as insect (e.g. *Nilaparvata lugens* (Choi et al. 2019; Park, Kwon, et al. 2019), *Laodelphax striatellus* (Park, Jung, et al. 2019; Seo, Jung, et al. 2019), *Spodoptera frugiperda* (Seo, Lee, et al. 2019), and *Aphis gossypii* (Bee et al. 2020; Park, Xi, et al. 2019)) and plant species (*Liriodendron tulipifera* (Park, Kim, et al. 2019)).

Sequence alignments of 10 conserved genes from 13 *Aspergillus*, one *Penicillium*, and one *Podospora* mitogenome (outgroup) were calculated by MAFFT v7.450 (Katoh and Standley 2013) and concatenated. The bootstrapped neighbor-joining and maximum likelihood phylogenetic trees were constructed using MEGA X (Kumar et al. 2018). Phylogenetic

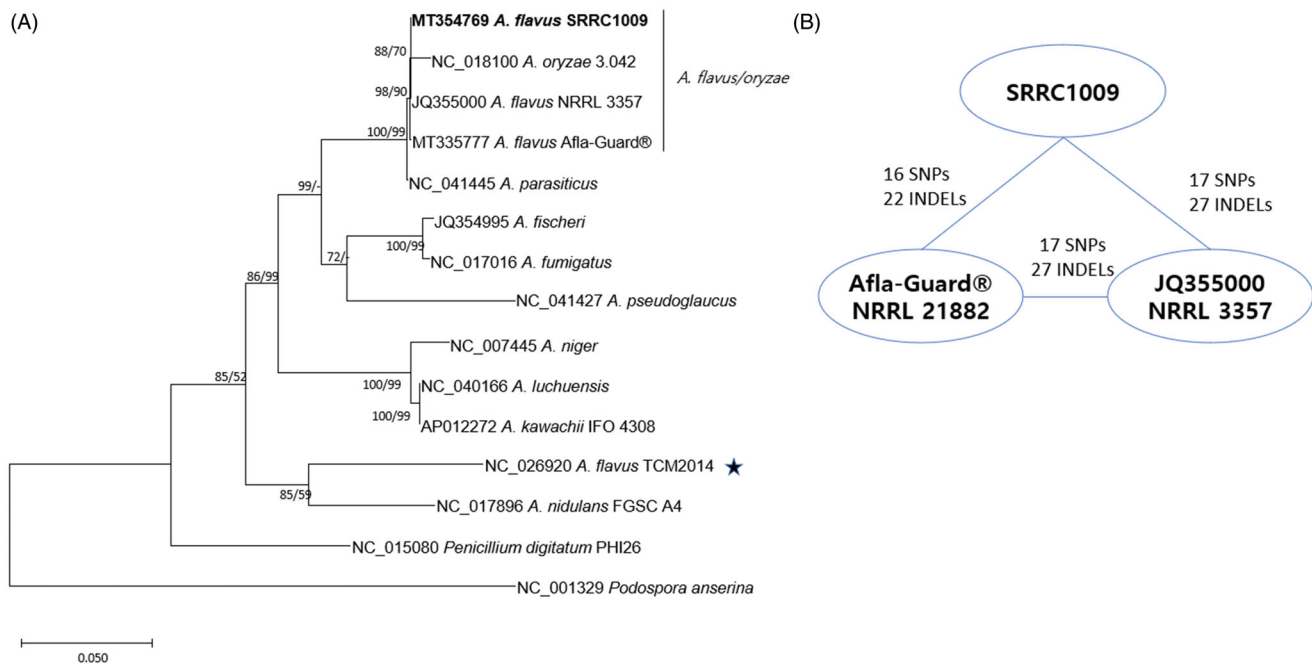


Figure 1. (A) Maximum likelihood (bootstrap repeat is 10,000) and neighbor joining (bootstrap repeat is 10,000) phylogenetic trees of 12 *Aspergillus*, one *Penicillium* mitochondrial genomes, and *Podospora* mitochondrial genome as an outgroup: *Aspergillus flavus* (MT354769 in this study, MT335777, JQ355000, and NC_026920), *Aspergillus oryzae* (NC_018100), *Aspergillus parasiticus* (NC_041445), *Aspergillus fischeri* (JQ354995), *Aspergillus fumigatus* (NC_017016), *Aspergillus pseudoglaucus* (NC_041427), *Aspergillus niger* (NC_007445), *Aspergillus kawachii* (AP012272), *Aspergillus luchuensis* (NC_040166), *Aspergillus nidulans* (NC_017896), *Penicillium digitatum* (NC_015080), and *Podospora anserina* (NC_001329). Phylogenetic tree was drawn based on maximum likelihood phylogenetic tree. The numbers above or below branches indicate bootstrap support values of maximum likelihood and neighbor joining phylogenetic trees, respectively. Star-marked *A. flavus* TCM2014 is not placed in *A. flavus/oryzae* group, indicating that it may be misidentified. (B) Summary of intraspecific variation of three *A. flavus* strains.

trees showed that three *A. flavus* mitogenomes were clustered in one clade together with *A. oryzae* mitogenome (Figure 1(A)).

Disclosure statement

No potential conflict of interest was reported by the author(s).

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Data availability statement

Mitochondrial genome sequence can be accessed via accession number MT354769 (<https://www.ncbi.nlm.nih.gov/nucore?term=MT354769>) in NCBI GenBank.

References

Bae Y, Park J., Lee W. 2020. The complete mitochondrial genome of *Aphis gossypii* Glover, 1877 (Hemiptera: Aphididae) isolated from *Plantago asiatica* in Korea. *Mitochondrial DNA Part B*. 5(3):2896–2898.

Choi NJ, Lee B-C, Park J, Park J. 2019. The complete mitochondrial genome of *Nilaparvata lugens* (Stål, 1854) captured in China (Hemiptera: Delphacidae): investigation of intraspecies variations between countries. *Mitochondrial DNA B*. 4(1):1677–1678.

Joardar V, Abrams NF, Hostetler J, Paukstelis PJ, Pakala S, Pakala SB, Zafar N, Abolude OO, Payne G, Andrianopoulos A, et al. 2012. Sequencing of mitochondrial genomes of nine *Aspergillus* and *Penicillium* species identifies mobile introns and accessory genes as main sources of genome size variability. *BMC Genomics*. 13(1):698.

Katoh K, Standley DM. 2013. MAFFT multiple sequence alignment software version 7: improvements in performance and usability. *Mol Biol Evol*. 30(4):772–780.

Klich MA, Pitt JI. 1988. Differentiation of *Aspergillus flavus* from *A. parasiticus* and other closely related species. *Trans Br Mycol Soc*. 91(1): 99–108.

Kumar S, Stecher G, Li M, Knyaz C, Tamura K. 2018. MEGA X: Molecular Evolutionary Genetics Analysis across Computing Platforms. *Mol Biol Evol*. 35(6):1547–1549.

Kwon W, Park J, Kim J-B, Park M-J, Kim T-S. 2019. Complete mitochondrial genome sequence of lettuce pathogenic fungus, *Fusarium oxysporum* f. sp. *lactucae* 16-086. *Mitochondrial DNA B*. 4(2):3227–3228.

Lee M-K, Park H-S, Han K-H, Hong S-B, Yu J-H. 2017. High molecular weight genomic DNA mini-prep for filamentous fungi. *Fungal Genet Biol*. 104:1–5.

Li H. 2013. Aligning sequence reads, clone sequences and assembly contigs with BWA-MEM. arXiv preprint arXiv:13033997.

Li H, Handsaker B, Wysoker A, Fennell T, Ruan J, Homer N, Marth G, Abecasis G, Durbin R. 2009. The sequence alignment/map format and SAMtools. *Bioinformatics*. 25(16):2078–2079.

Nierman WC, Yu J, Fedorova-Abrams ND, Losada L, Cleveland TE, Bhatnagar D, Bennett JW, Dean R, Payne GA. 2015. Genome sequence of *Aspergillus flavus* NRRL 3357, a strain that causes aflatoxin contamination of food and feed. *Genome Announc*. 3(2):e00168–15.

Park J, Jung JK, Ho Koh Y, Park J, Seo BY. 2019. The complete mitochondrial genome of *Laodelphax striatellus* (Fallén, 1826) (Hemiptera: Delphacidae) collected in a mid-Western part of Korean peninsula. *Mitochondrial DNA B*. 4(2):2229–2230.

- Park J, Kim Y, Kwon M. 2019. The complete mitochondrial genome of tulip tree, *Liriodendron tulipifera* L. (Magnoliaceae): intra-species variations on mitochondrial genome. *Mitochondrial DNA B*. 4(1):1308–1309.
- Park J, Kwon W, Park J, Kim H-J, Lee B-C, Kim Y, Choi NJ. 2019. The complete mitochondrial genome of *Nilaparvata lugens* (stål, 1854) captured in Korea (Hemiptera: Delphacidae). *Mitochondrial DNA B*. 4(1):1674–1676.
- Park J, Xi H, Kim Y, Park J, Lee W. 2019. The complete mitochondrial genome of *Aphis gossypii* Glover, 1877 (Hemiptera: Aphididae) collected in Korean peninsula. *Mitochondrial DNA B*. 4(2):3007–3009.
- Park et al. Under review. Comprehensive analysis *Aspergillus* and *Penicillium* mitochondrial genomes reveals conserved and divergent features of the species. <https://doi.org/10.1080/23802359.2020.1825129>
- Payne G, Nierman W, Wortman J, Pritchard B, Brown D, Dean R, Bhatnagar D, Cleveland T, Machida M, Yu J. 2006. Whole genome comparison of *Aspergillus flavus* and *A. oryzae*. *Med Mycol*. 44(Suppl. 1):S9–S11.
- Seo BY, Jung JK, Ho Koh Y, Park J. 2019. The complete mitochondrial genome of *Laodelphax striatellus* (Fallén, 1826) (Hemiptera: Delphacidae) collected in a southern part of Korean peninsula. *Mitochondrial DNA B*. 4(2):2242–2243.
- Seo BY, Lee G-S, Park J, Xi H, Lee H, Lee J, Park J, Lee W. 2019. The complete mitochondrial genome of the fall armyworm, *Spodoptera frugiperda* Smith, 1797 (Lepidoptera; Noctuidae), firstly collected in Korea. *Mitochondrial DNA B*. 4(2):3918–3920.
- Squire RA. 1981. Ranking animal carcinogens: a proposed regulatory approach. *Science*. 214(4523):877–880.
- Weaver MA, Scheffler BE, Duke M, Ballard L, Abbas HK, Grodowitz MJ. 2017. Genome sequences of three strains of *Aspergillus flavus* for the biological control of aflatoxin. *Genome Announc*. 5(44):e01204–17.
- Yan Z, Chen D, Shen Y, Ye B. 2016. The complete mitochondrial genome sequence of *Aspergillus flavus*. *Mitochondrial DNA A*. 27(4):2671–2672.
- Zerbino DR, Birney E. 2008. Velvet: algorithms for *de novo* short read assembly using de Bruijn graphs. *Genome Res*. 18(5):821–829.
- Zhao Q-Y, Wang Y, Kong Y-M, Luo D, Li X, Hao P. 2011. Optimizing *de novo* transcriptome assembly from short-read RNA-Seq data: a comparative study. *BMC Bioinf*. 12(Suppl. 14):S2.