Characterization and phylogenetic analysis of the complete mitochondrial genome of pathogen *Trichosporon inkin* (Trichosporonales: Trichosporonaceae)

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**ABSTRACT**

In the present study, the complete mitochondrial genome of *Trichosporon inkin* was sequenced and assembled. The complete mitochondrial genome of *T. inkin* contained 22 protein-coding genes (PCG), 2 ribosomal RNA (rRNA) genes, and 24 transfer RNA (tRNA) genes. The total size of the *T. inkin* mitochondrial genome is 39,466 bp, with the GC content of 27.56%. Phylogenetic analysis based on combined mitochondrial gene dataset indicated that the *T. inkin* exhibited a close relationship with *Trichosporon asahii*.

The genus *Trichosporon* is a group of yeast-like fungi, living widespread in nature (Marine et al. 2015; Duarte-Oliveira et al. 2017). Over 50 species have been assigned into this genus (Colombo et al. 2011; Gouba et al. 2014). Most species from this genus are classical opportunistic pathogens, which reside harmlessly as commensals on the skin and the gastrointestinal tract of healthy individuals (Zhang et al. 2011; Guo et al. 2018). Species identification of *Trichosporon* can be used to trace the origin of nosocomial infection and the hospital or regional specificity of isolated strains (Sun et al. 2012; Guo et al. 2018). Mitochondrial genotypes have been widely used in the phylogenetic analysis of basidiomycete species (Li et al. 2019b; Li, He, et al. 2020; Wang, Song, et al. 2020). However, up to now, only one complete mitochondrial genome from the genus *Trichosporon* has been reported (*Trichosporon asahii*). The mitochondrial genome of *Trichosporon inkin* will promote the understanding of the phylogeny, origin, and taxonomy of this important genus.

The specimen (*T. inkin*) was collected from a Hospital in Sichuan, China (104.05 E; 30.65 N) in 2017. The *T. inkin* was isolated from the cultures of diseased foot tissue and identified by deoxyribonucleic acid sequencing. We stored the specimen in Culture Collection Center of Chengdu Medical College (No. Trisp_x3). The complete mitochondrial genome of *T. inkin* was sequenced and de novo assembled according to previously described methods (Li, Liao, et al. 2018; Li, Ren, et al. 2020). Briefly, the protein-coding genes, rRNA genes, tRNA genes, and introns of the *T. inkin* mitogenome were initially annotated using MITOS (Bernt et al. 2013) and MFannot (Valach et al. 2014), both based on the genetic code 4. The PCGs and rRNA genes were further annotated according to methods described previously (Wang, Jia, et al. 2020; Wang, Wang, et al. 2020; Wu et al. 2021). The tRNA genes in the *T. inkin* mitogenome were also predicted with tRNAscan-SE v1.3.1 (Lowe and Chan 2016). Introns in the *T. inkin* mitogenome were named according to previous studies (Zhang and Zhang 2019). The complete mitochondrial genome of *T. inkin* was stored in Chengdu Medical College (No. DNA_Trisp_x3). Sequencing libraries were constructed using a NEBNext<sup>®</sup> Ultra<sup>TM</sup> II DNA Library Prep Kit (NEB, Beijing, China). The Illumina HiSeq 2500 Platform (Illumina, San Diego, CA) was used to conduct whole genomic sequencing (WGS) of *T. inkin*. The mitochondrial genome of *T. inkin* was de novo assembled using SPAdes 3.9.0 (Bankevich et al. 2012; Li, Ren, et al. 2020) with a kmer size of 17. Since organelle sequences usually have more copies than nuclear gene sequences, the mitochondrial genome we finally obtained showed high coverage. In addition, MITObim (Hahn et al. 2013), and NOVOPlasty (Dierckxsens et al. 2017) were also used to test the assembly of this study. All the software obtained complete mitogenomes identical to this study, which proves that the mitochondrial genome obtained in the present study is reliable.

We annotated the complete mitochondrial genome of *T. inkin* according to previously described methods (Li, Chen, et al. 2018; Li, Wang, et al. 2018). Briefly, the protein-coding genes, rRNA genes, tRNA genes, and introns of the *T. inkin* mitogenome were initially annotated using MITOS (Bernt et al. 2013) and MFannot (Valach et al. 2014), both based on the genetic code 4. The PCGs and rRNA genes were further annotated according to methods described previously (Wang, Jia, et al. 2020; Wang, Wang, et al. 2020; Wu et al. 2021). The tRNA genes in the *T. inkin* mitogenome were also predicted with tRNAscan-SE v1.3.1 (Lowe and Chan 2016). Introns in the *T. inkin* mitogenome were named according to previous studies (Zhang and Zhang 2019).

The complete mitochondrial genome of *T. inkin* is 39,466 bp in length. The base composition of the *T. inkin* mitogenome is as follows: A (36.17%), T (36.27%), G (13.99%), and C (13.57%). The complete mitochondrial genome of *T. inkin* was deposited in GenBank under the accession number MZ770610.
T. inkin contains 22 protein-coding genes, 2 ribosomal RNA genes (rns and rnl), and 24 transfer RNA genes. Nine introns were detected in the T. inkin mitogenome, including 4 in the cox1, 2 in cox2, 1 in cob, nad5, and rnl genes, respectively. These introns all belonged to the Group I. Introns in PCGs of the T. inkin mitogenome were named according to previous studies (Zhang and Zhang 2019), including Tin.cox1P386, Tin.cox1P709, Tin.cox1P807, Tin.cox1P867, Tin.cox2P318, Tin.nad5P717, and Tin.cobP506. Six intronic ORFs were detected in the T. inkin mitogenome, which encoded LAGLIDADG endonucleases. We constructed a phylogenetic tree for 18 basidiomycete species to investigate the phylogenetic status of T. inkin. Rhizopogon salebrosus from the order Boletales was set as an outgroup (Li, Ren, et al. 2019). The Bayesian analysis (BI) method was used to construct phylogenetic tree based on the combined 14 core protein-coding genes according to previously described methods (Li, Wang, et al. 2019; Li et al. 2019a; Li, Yang, et al. 2020). As shown in the phylogenetic tree (Figure 1), the mitochondrial genome of T. inkin exhibited a close relationship with that of T. asahii (JH925097).

**Disclosure statement**

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

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**Data availability statement**

This mitogenome of Trichosporon inkin was submitted to GenBank under the accession number of MT801082. [https://www.ncbi.nlm.nih.gov/nuccore/ MT801082].

**Figure 1.** Bayesian phylogenetic analysis of 18 species based on the combined 14 core protein-coding genes. Support values are Bayesian posterior probabilities (BPP). Accession numbers of mitochondrial sequences used in the phylogenetic analysis are listed in brackets after species.

**References**

Bankevich A, Nurk S, Antipov D, Gurevich AA, Dvorkin M, Kulikov AS, Lesin VM, Nikolenko SI, Pham S, Pyshkin AV, et al. 2012. SPAdes: a new genome assembly algorithm and its applications to single-cell sequencing. J Comput Biol. 19(5):455–477.

Bernt M, Donath A, Juhling F, Externbrink F, Florentz C, Fritzsch G, Putz J, Middendorf M, Standler PF. 2013. MITOS: improved de novo metazoan mitochondrial genome annotation. Mol Phylogenet Evol. 69(2):313–319.

Colombo AL, Padovan AC, Chaves GM. 2011. Current knowledge of Trichosporon spp. and Trichosporonosis. Clin Microbiol Rev. 24(4):682–700.

Dierckxsens N, Mardulyn P, Smits G. 2017. NOVOPlasty: de novo assembly of organelle genomes from whole genome data. Nucleic Acids Res. 45(4):e18.

Duarte-Oliveira C, Rodrigues F, Goncalves SM, Goldman GH, Carvalho A, Cunha C. 2017. The cell biology of the Trichosporon-host interaction. Front Cell Infect Microbiol. 7:118.

Gouba N, Raoult D, Drancourt M. 2014. Eukaryote culturomics of the gut reveals new species. PLoS One. 9(9):e106994.

Guo LN, Yu SY, Hsueh PR, Al-Hatmi AMS, Meis JF, Hagen F, Xiao M, Wang H, Barresi C, Zhou ML, et al. 2018. Invasive infections due to Trichosporon: species distribution, genotyping, and antifungal susceptibilities from a multicenter study in China. J Clin Microbiol. 57(2):e01505.

Hahn C, Bachmann L, Chevreux B. 2013. Reconstructing mitochondrial genomes directly from genomic next-generation sequencing reads—a baiting and iterative mapping approach. Nucleic Acids Research. 41(13):e129.

Li Q, Chen C, Xiong C, Jin X, Chen Z, Huang W. 2018. Comparative mitogenomics reveals large-scale gene rearrangements in the mitochondrial genome of two Pleurotus species. Appl Microbiol Biotechnol. 102(14):6143–6153.

Li Q, He X, Ren Y, Xiong C, Jin X, Peng L, Huang W. 2020. Comparative mitogenome analysis reveals mitochondrial genome differentiation in ectomycorrhizal and asymbiotic Amanita species. Front Microbiol. 11:1382.

Li Q, Liao M, Yang M, Xiong C, Jin X, Chen Z, Huang W. 2018. Characterization of the mitochondrial genomes of three species in the ectomycorrhizal genus Cantharellus and phylogeny of Agaricomycetes. Int J Biol Macromol. 118:756–769.

Li Q, Ren Y, Shi X, Peng L, Zhao J, Song Y, Zhao G. 2019. Comparative mitochondrial genome analysis of two ectomycorrhizal fungi (Rhizopogon) reveals dynamic changes of intron and phylogenetic
relationships of the subphylum Agaricomycotina. Int J Mol Sci. 20(20): 5167.

Li Q, Ren Y, Xiang D, Shi X, Zhao J, Peng L, Zhao G. 2020. Comparative mitogenome analysis of two ectomycorrhizal fungi (Paxillus) reveals gene rearrangement, intron dynamics, and phylogeny of basidiomycetes. IMA Fungus. 11:12.

Li Q, Wang Q, Chen C, Jin X, Chen Z, Xiong C, Li P, Zhao J, Huang W. 2018. Characterization and comparative mitogenomic analysis of six newly sequenced mitochondrial genomes from ectomycorrhizal fungi (Russula) and phylogenetic analysis of the Agaricomycetes. Int J Biol Macromol. 119:792–802.

Li Q, Wang Q, Jin X, Chen Z, Xiong C, Li P, Liu Q, Huang W. 2019. Characterization and comparative analysis of six complete mitochondrial genomes from ectomycorrhizal fungi of the Lactarius genus and phylogenetic analysis of the Agaricomycetes. Int J Biol Macromol. 121: 249–260.

Li Q, Wang Q, Jin X, Chen Z, Xiong C, Li P, Zhao J, Huang W. 2019a. Characterization and comparison of the mitochondrial genomes from two Lyophyllum fungal species and insights into phylogeny of Agaricomycetes. Int J Biol Macromol. 121:364–372.

Li Q, Wang Q, Jin X, Chen Z, Xiong C, Li P, Zhao J, Huang W. 2019b. The first complete mitochondrial genome from the family Hygrophoraceae (Hygrophorus russula) by next-generation sequencing and phylogenetic implications. Int J Biol Macromol. 122:1313–1320.

Li Q, Xiang D, Wan Y, Wu Q, Wu X, Ma C, Song Y, Zhao G, Huang W. 2019. The complete mitochondrial genomes of five important medicinal Ganoderma species: Features, evolution, and phylogeny. Int J Biol Macromol. 139:397–408.

Li Q, Yang L, Xiang D, Wan Y, Wu Q, Huang W, Zhao G. 2020. The complete mitochondrial genomes of two model ectomycorrhizal fungi (Laccaria): features, intron dynamics and phylogenetic implications. Int J Biol Macromol. 145:974–984.

Lowe TM, Chan PP. 2016. tRNAscan-SE On-line: integrating search and context for analysis of transfer RNA genes. Nucleic Acids Res. 44(W1): W54–W57.

Marine M, Brown NA, Riano-Pachon DM, Goldman GH. 2015. On and under the skin: emerging basidiomycetous yeast infections caused by Trichosporon species. PloS Pathog. 11(7):e1004982.

Sun W, Su J, Xu S, Yan D. 2012. Trichosporon asahii causing nosocomial urinary tract infections in intensive care unit patients: genotypes, virulence factors and antifungal susceptibility testing. J Med Microbiol. 61(12):1750–1757.

Valach M, Burger G, Gray MW, Lang BF. 2014. Widespread occurrence of organelle genome-encoded 5S rRNAs including permuted molecules. Nucleic Acids Res. 42(22):13764–13777.

Wang X, Jia LH, Wang MD, Yang H, Chen MY, Li X, Liu HY, Li Q, Liu N. 2020. The complete mitochondrial genome of medicinal fungus Taiwanofungus camphoratus reveals gene rearrangements and intron dynamics of Polyporales. Sci Rep. 10(1):16500.

Wang X, Song A, Wang F, Chen M, Li X, Li Q, Liu N. 2020. The 206 kbp mitochondrial genome of Phanerochaete carnosa reveals dynamics of introns, accumulation of repeat sequences and plasmid-derived genes. Int J Biol Macromol. 162:209–219.

Wang X, Wang YJ, Yao W, Shen JW, Chen MY, Gao M, Ren JN, Li Q, Liu N. 2020. The 256 kb mitochondrial genome of Clavaria fumosa is the largest among phylum Basidiomycota and is rich in introns and intronic ORFs. IMA Fungus. 11(1):7.

Wu P, Bao Z, Tu W, Li L, Xiong C, Li P, Gui M, Huang W, Li Q. 2021. The mitogenomes of two saprophytic Boletales species (Coniophora) reveals intron dynamics and accumulation of plasmid-derived and non-conserved genes. Comput Struct Biotechnol J. 19:401–414.

Zhang E, Sugita T, Tsuboi R, Yamazaki T, Makimura K. 2011. The opportunistic yeast pathogen Trichosporon asahii colonizes the skin of healthy individuals: analysis of 380 healthy individuals by age and gender using a nested polymerase chain reaction assay. Microbiol Immunol. 55(7):483–488.

Zhang S, Zhang YJ. 2019. Proposal of a new nomenclature for introns in protein-coding genes in fungal mitogenomes. IMA Fungus. 10(15):15.