

Complete Mitochondrial Genome Sequence of *Lichtheimia ramosa* (syn. *Lichtheimia hongkongensis*)

Shui-Yee Leung,^a Yi Huang,^a Susanna K. P. Lau,^{a,b,c,d} Patrick C. Y. Woo^{a,b,c,d}

Department of Microbiology,^a State Key Laboratory of Emerging Infectious Diseases,^b Research Centre of Infection and Immunology,^c and Carol Yu Centre of Infection,^d The University of Hong Kong, Hong Kong

We report the complete mitochondrial genome sequence of *Lichtheimia ramosa* (syn. *Lichtheimia hongkongensis*), the first complete mitochondrial DNA sequence of the genus *Lichtheimia*. This 31.8-kb mitochondrial genome encodes 11 subunits of respiratory chain complexes, 3 ATP synthase subunits, 25 tRNAs, and small and large rRNAs, with the gene order *atp9-cox2-atp6-cox3-cox1-nad2-nad3-cob-nad1-nad6-nad5-nad4l-nad4-atp8*.

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Address correspondence to Susanna K. P. Lau, skplau@hkucc.hku.hk, or Patrick C. Y. Woo, pcywoo@hkucc.hku.hk.

Mucormycosis is an opportunistic infection caused by molds of the order *Mucorales*. The most common species of *Mucorales* associated with mucormycosis belong to the genera *Rhizopus*, *Rhizomucor*, *Mucor*, and *Absidia*. Among the *Absidia* species, the most important species associated with mucormycosis is *Absidia corymbifera*. According to physiological, phylogenetic, and morphological data, it was proposed that three *Absidia* species, *A. corymbifera*, *A. blakesleeana*, and *A. hyalospora*, should be reclassified as a separate family, *Lichtheimiaceae* fam. nov., and the three species renamed *Lichtheimia corymbifera*, *Lichtheimia blakesleeana*, and *Lichtheimia hyalospora* (1, 2). *L. blakesleeana* was subsequently reduced to a synonym of *L. hyalospora* (3). Recently, we and others described a novel *Lichtheimia* species, *Lichtheimia ramosa* (syn. *Lichtheimia hongkongensis*), from patients with mucormycosis (4, 5). We also found that a significant number of reported *L. corymbifera* infections were actually caused by *L. ramosa* (6). Among the species of *Mucorales*, the only complete mitochondrial genome sequenced is that of *Rhizopus oryzae* (7). In this article, we report the complete mitochondrial genome sequence of *L. ramosa* HKU22.

Mitochondrial DNA (mtDNA) of *L. ramosa* was purified using the mitochondrial DNA isolation kit, according to the manufacturer's instructions (PromoKine), and sequenced using random clone libraries, with the nebulized mitochondrial DNA cloned into a modified pBlueScript KS+ vector. Both DNA strands were sequenced by the dideoxy chain-termination method and assembled using CAP3 (<http://pbil.univ-lyon1.fr/cap3.php>) (8). The putative open reading frames (ORFs) were denoted using Artemis, with the genetic code of mold. Genes were assigned functions through a BLASTp search against fungal mitochondrion-encoded proteins available in the GenBank database. Introns and rRNAs were identified by a BLASTn pairwise comparison of *L. ramosa* mtDNA with other fungal mitochondrial DNAs. The BLASTn results were viewed through ACT, a DNA sequence comparison viewer based on Artemis, and the exon/intron boundaries were adjusted manually. tRNAs were predicted by tRNAscan-SE 1.21

(<http://lowelab.ucsc.edu/tRNAscan-SE/>) (9). The core structures of the group I introns were inferred by the program RNAweasel (<http://megasun.bch.umontreal.ca/RNAweasel/>).

The mtDNA of *L. ramosa* is a circular DNA molecule of 31,830 bp. The G+C content is 34.1%. Genes are carried by both DNA strands, with the gene order *atp9-cox2-atp6-cox3-cox1-nad2-nad3-cob-nad1-nad6-nad5-nad4l-nad4-atp8*. Structural genes occupy 57.9% of the genome (39.1% protein-coding exons, 5.8% tRNA genes, and 13.0% rRNA genes), 8.0% is occupied by intergenic spacers, and 1.9% is occupied by introns. The genome contains 11 genes encoding subunits of respiratory chain complexes (cytochrome oxidase subunits I, II, and III [*cox1*, *cox2*, and *cox3*, respectively], apocytochrome b [*cob*], and the reduced nicotinamide adenine dinucleotide ubiquinone oxidoreductase subunits [*nad1*, *nad2*, *nad3*, *nad4*, *nad4l*, *nad5*, and *nad6*]) and three genes encoding ATP synthase subunits (*atp6*, *atp8*, and *atp9*). It also encodes 25 tRNAs and the 23S and 16S rRNAs of the large and small subunits of the ribosome (*rnl* and *rns*, respectively). There is one 120-bp ORF, with an unknown function, between *nad4* and *atp8*. The *cox1* and *cox2* genes contain one group I intron each. Phylogenetically, the mtDNA of *L. ramosa* is most closely related to that of *R. oryzae*.

Nucleotide sequence accession number. The mtDNA sequence is deposited in GenBank with accession no. [KJ561171](https://www.ncbi.nlm.nih.gov/nuccore/KJ561171).

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REFERENCES

- Hoffmann K, Discher S, Voigt K. 2007. Revision of the genus *Absidia* (*Mucorales*, *Zygomycetes*) based on physiological, phylogenetic, and morphological characters; thermotolerant *Absidia* spp. form a coherent group, *Mycocladiaceae* fam. nov. *Mycol. Res.* 111:1169–1183. <http://dx.doi.org/10.1016/j.mycres.2007.07.002>.
- Hoffmann K, Walther G, Voigt K. 2009. *Mycocladius* vs. *Lichtheimia*: a correction (*Lichtheimiaceae* fam. nov., *Mucorales*, *Mucoromycotina*). *Mycol. Res.* 113:275–278. <http://dx.doi.org/10.1016/j.mycres.2009.02.001>.

3. Alastruey-Izquierdo A, Hoffmann K, de Hoog GS, Rodriguez-Tudela JL, Voigt K, Bibashi E, Walther G. 2010. Species recognition and clinical relevance of the zygomycetous genus *Lichtheimia* (syn. *Absidia pro parte, Mycocladus*). *J. Clin. Microbiol.* 48:2154–2170. <http://dx.doi.org/10.1128/JCM.01744-09>.
4. Garcia-Hermoso D, Hoinard D, Gantier JC, Grenouillet F, Dromer F, Dannaoui E. 2009. Molecular and phenotypic evaluation of *Lichtheimia corymbifera* (formerly *Absidia corymbifera*) complex isolates associated with human mucormycosis: rehabilitation of *L. ramosa*. *J. Clin. Microbiol.* 47:3862–3870. <http://dx.doi.org/10.1128/JCM.02094-08>.
5. Woo PC, Lau SK, Ngan AH, Tung ET, Leung SY, To KK, Cheng VC, Yuen KY. 2010. *Lichtheimia hongkongensis* sp. nov., a novel *lichtheimia* spp. associated with rhinocerebral, gastrointestinal, and cutaneous mucormycosis. *Diagn. Microbiol. Infect. Dis.* 66:274–284. <http://dx.doi.org/10.1016/j.diagmicrobio.2009.10.009>.
6. Woo PCY, Leung SY, Ngan AHY, Lau SKP, Yuen KY. 2012. A significant number of reported *Absidia corymbifera* (*Lichtheimia corymbifera*) infections are caused by *Lichtheimia ramosa* (syn. *Lichtheimia hongkongensis*): an emerging cause of mucormycosis. *Emerg. Microbes Infect.* 1:e15. <http://dx.doi.org/10.1038/emi.2012.11>.
7. Seif E, Leigh J, Liu Y, Roewer I, Forget L, Lang BF. 2005. Comparative mitochondrial genomics in zygomycetes: bacteria-like RNase P RNAs, mobile elements and a close source of the group I intron invasion in angiosperms. *Nucleic Acids Res.* 33:734–744. <http://dx.doi.org/10.1093/nar/gki199>.
8. Huang X, Madan A. 1999. CAP3: a DNA sequence assembly program. *Genome Res.* 9:868–877. <http://dx.doi.org/10.1101/gr.9.9.868>.
9. Lowe TM, Eddy SR. 1997. tRNAscan-SE: a program for improved detection of transfer RNA genes in genomic Sequence. *Nucleic Acids Res.* 25:0955–0964. <http://dx.doi.org/10.1093/nar/25.5.955>.