

# Draft Genome Sequence of *Mycobacterium bolletii* Strain M24, a Rapidly Growing Mycobacterium of Contentious Taxonomic Status

Yan Ling Wong,<sup>a</sup> Siew Woh Choo,<sup>b</sup> Joon Liang Tan,<sup>b,c</sup> Chia Sui Ong,<sup>c</sup> Kee Peng Ng,<sup>a</sup> and Yun Fong Ngeow<sup>a</sup>

Department of Medical Microbiology, Faculty of Medicine, University of Malaya, Kuala Lumpur, Malaysia<sup>a</sup>; Dental Research and Training Unit, Faculty of Dentistry, University of Malaya, Kuala Lumpur, Malaysia<sup>b</sup>; and Faculty of Information Science and Technology, Multimedia University, Melaka, Malaysia<sup>c</sup>

**The whole-genome sequence of *Mycobacterium bolletii* M24, isolated from the bronchoalveolar lavage fluid of a Malaysian patient, is reported here. The circular chromosome of 5,507,730 bp helped to clarify the taxonomic position of this organism within the *M. abscessus* complex and revealed the presence of proteins potentially important for pathogenicity in a human host.**

Among the rapidly growing atypical mycobacteria, *Mycobacterium abscessus* is the species most frequently associated with human infections. It is a member of the *M. chelonae*-*M. abscessus* complex that comprises many closely related genotypes. Since its first description by Moore and Frerichs in 1953 (6), this species has undergone many changes in taxonomic status owing to the ambiguous results of molecular differentiation. Adekambi et al. (1, 2) suggested the creation of three species to include *M. abscessus sensu stricto*, *M. bolletii*, and *M. massiliense*. Other workers recommended the reclassification of the three species into a single species of *M. abscessus* (5), and recently Leão et al. combined *M. bolletii* and *M. massiliense* into one subspecies named *M. abscessus* subspecies *bolletii* comb. nov., leaving other members in a second subspecies named *M. abscessus* subspecies *abscessus* (4). With the availability of whole genome sequences, it is anticipated that more members of this group of bacteria will be described that may necessitate further changes in nomenclature.

The genome of *M. bolletii* strain M24 was sequenced using the Illumina GA 2× sequencing platform and annotated with the Rapid Annotation Subsystem Technology (RAST) pipeline (3). The 12,215,918 Illumina sequencing reads were assembled using the Genomics Workbench 4.9, resulting in 281 contigs with a genome size of 5,507,730 bp. Using the RAST server, 5,605 predicted coding sequences were identified. In addition, 3 rRNAs and 86 tRNAs were predicted. A phylogenetic tree based on the RNA polymerase beta subunit (*rpoB*) gene was constructed by using MEGA (Molecular Evolutionary Genetic Analysis). In comparison with reference strains of *M. bolletii* (CIP 108541, FI-09284, P52), *M. abscessus sensu stricto* (PCH-017, FI-08197, ATCC 19977), and *M. massiliense* (CRM642, CRM552, IAL035), M24 was identified as *M. bolletii* by its clustering with the three reference strains of *M. bolletii*. Pairwise alignments of M24 to all reference sequences using ClustalW showed that M24 is 95% identical to all reference strains of *M. abscessus sensu stricto*, 98% identical to the three reference strains of *M. massiliense*, and 99% to 100% identical to the reference strains of *M. bolletii*.

Of the 5,605 predicted coding sequences, 1,585 (29%) are involved in 377 subsystems. The highest count (425 genes) is for sequences involved with amino acids and derivatives. The lowest count (1 gene) is in the motility and chemotaxis subsystem category. There are 34 genes encoding gene products that may be involved with virulence, disease, and defense, of which 21 are linked with resistance to antibiotics and toxic compounds and 13

with invasion and intracellular resistance. Further analysis is ongoing to identify putative proteins associated with the pathogenicity of this organism in the human host.

**Nucleotide sequence accession numbers.** The *M. bolletii* strain M24 genome sequence and annotation data have been deposited in NCBI GenBank under the accession number [AJLY000000000](https://www.ncbi.nlm.nih.gov/nuccore/AJLY000000000). The version described in this paper is the first version, AJLY01000000.

## ACKNOWLEDGMENTS

This work was supported by research grants UM.C/HIR/MOHE/08 and UM.C/625/1/HIR/004 from the University of Malaya, Kuala Lumpur, Malaysia.

## REFERENCES

- Adekambi T, et al. 2004. Amoebal coculture of “*Mycobacterium massiliense*” sp. nov. from the sputum of a patient with hemoptoic pneumonia. *J. Clin. Microbiol.* 42:5493–5501.
- Adekambi T, Berger P, Raoult D, Drancourt M. 2006. *rpoB* gene sequence-based characterization of emerging non-tuberculous mycobacteria with descriptions of *Mycobacterium bolletii* sp. nov., *Mycobacterium phocaticum* sp. nov. and *Mycobacterium aubagnense* sp. nov. *Int. J. Syst. Evol. Microbiol.* 56(Pt 1):133–143.
- Aziz RK, et al. 2008. The RAST Server: Rapid Annotations using Subsystems Technology. *BMC Genomics* 9:75.
- Leão SC, Tortoli E, Euzéby JP, Garcia MJ. 2011. Proposal that *Mycobacterium massiliense* and *Mycobacterium bolletii* be united and reclassified as *Mycobacterium abscessus* subsp. *bolletii* comb. nov., designation of *Mycobacterium abscessus* subsp. *abscessus* subsp. nov. and emended description of *Mycobacterium abscessus*. *Int. J. Syst. Evol. Microbiol.* 61(Pt 9):2311–2313.
- Macheras E, et al. 2011. Multilocus sequence analysis and *rpoB* sequencing of *Mycobacterium abscessus* (sensu lato) strains. *J. Clin. Microbiol.* 49:491–499.
- Moore M, Frerichs JB. 1953. An unusual acid-fast infection of the knee with subcutaneous, abscess-like lesions of the gluteal region; report of a case with a study of the organism, *Mycobacterium abscessus*, n. sp. *J. Invest. Dermatol.* 20:133–169.

Received 25 May 2012 Accepted 5 June 2012

Address correspondence to Siew Woh Choo, [lchoo@um.edu.my](mailto:lchoo@um.edu.my), or Yun Fong Ngeow, [yunngeow@um.edu.my](mailto:yunngeow@um.edu.my).

Y.L.W. and S.W.C. contributed equally to this project and should be considered co-first authors.

Copyright © 2012, American Society for Microbiology. All Rights Reserved.

doi:10.1128/JB.00916-12