

Annotated Genome Sequence of *Mycobacterium massiliense* Strain M154, Belonging to the Recently Created Taxon *Mycobacterium abscessus* subsp. *bolletii* comb. nov.

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Mycobacterium massiliense has recently been proposed as a member of *Mycobacterium abscessus* subsp. *bolletii* comb. nov. Strain M154, a clinical isolate from the bronchoalveolar lavage fluid of a Malaysian patient presenting with lower respiratory tract infection, was subjected to shotgun DNA sequencing with the Illumina sequencing technology to obtain whole-genome sequence data for comparison with other genetically related strains within the *M. abscessus* species complex.

The importance of nontuberculous mycobacteria as human pathogens has been increasingly recognized in the past few decades. With improved detection methods, particularly those involving molecular analyses, new species are being identified in an expanding range of clinical material (3, 4). There are controversies, however, in the classification of these acid-fast bacteria.

Mycobacterium massiliense, a nonpigmented fast grower, was first identified as a species within the *M. abscessus* complex (1). Subsequently, it was reclassified together with *M. bolletii* into a subspecies named *M. abscessus* subspecies *bolletii* comb. nov (6). With the appearance of increasing numbers of genetically closely related members in the *M. abscessus* species, differentiation by 16S rRNA gene sequencing is no longer adequate (7). Alternative and more sophisticated sequencing platforms are needed to clarify the correct taxonomic positions of mycobacteria within the *M. abscessus* complex.

The *ermA1* gene has been shown to be a suitable marker for the differentiation of *M. massiliense* from other rapidly growing mycobacteria (5). M154 was identified as *M. massiliense* based on the size of its *ermA1* gene sequence—which was identical with that of *M. massiliense* strain RGM134 but different from those of *M. abscessus* strain ATCC 19977 and *M. bolletii* strain KCTC 19281—as well as the presence of a truncated region in the *ermA1* sequence in M154 which was not present in the other two subspecies (5).

The genome of *M. massiliense* M154 was sequenced by using the Illumina GA 2X technology, which generated 9,920,574 reads. The sequences obtained were assembled with a commercial software, CLCBio Genomics Workbench 4.9, resulting in 44 contigs with the quality measurement of N25, contig size of 627,736, N50 contig size of 307,107 bp, and N75 contig size of 183,674 bp.

The resulting contigs were annotated using the Rapid Annotation Subsystem Technology pipeline (2). The draft genome sequence shows a genome size of 4,801,011 bp. The automated pipeline identified 4,706 predicted coding sequences, with G+C content of 64.2%. There are 1,506 (33% of the total) predicted coding sequences in the 375 RAST subsystems, and 45 tRNA and 3 rRNA were identified. Comparative analysis showed the top five closest neighbors of M154 to be *M. abscessus*, *M. smegmatis* strain MC2 155, *Mycobacterium* sp. strain MCS, *Mycobacterium* sp. strain JLS, and *Mycobacterium* sp. strain KMS, with scores of 345 to 508. These closest neighbors were achieved by blasting M154

putative genes into FIGfams, based on the isofunctional homolog property (2).

Nucleotide sequence accession numbers. The *M. massiliense* strain M154 genome sequence and annotation data have been deposited in NCBI GenBank under the accession number [AJMA00000000](https://doi.org/10.1093/aaj/aak000). The version described in this paper is the first version, AJMA01000000.

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