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Genomic Analysis of Mycobacterium abscessus Strain M139, Which Has an Ambiguous Subspecies Taxonomic Position

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Mycobacterium abscessus is a ubiquitous, rapidly growing species of nontuberculous mycobacteria that colonizes organic surfaces and is frequently associated with opportunistic infections in humans. We report here the draft genome sequence of Mycobacterium abscessus strain M139, which shows genomic features reported to be characteristic of both Mycobacterium abscessus subsp. abscessus and Mycobacterium abscessus subsp. massiliense.

The nontuberculous mycobacteria (NTM) are gaining importance as human pathogens, as they are being recovered from an increasingly wide spectrum of clinical material (2, 6, 9). Many of them show intrinsic resistance to antituberculosis drugs currently in use, and their identification beyond the species level is sometimes necessary to predict their response to antibiotic therapy (3). The species Mycobacterium abscessus, which has been associated with skin and soft tissue lesions, as well as pulmonary and other deep-seated sepsis (5), has been subdivided into subspecies by DNA sequence variation detected by molecular tests based on individual genes or a combination of genes (7). We had shotgun sequenced another genome for this species, from strain M139 isolated from the sputum sample of a 26-year-old Nepalese male presenting with hemoptysis using the Illumina GA 2X technology. With this technology, we generated 4,086,927 sequencing reads. These sequences were processed and assembled with Genomics Workbench 4.9, resulting in 42 contigs (36 contigs have a genomic size of >500 bp) with an N50, contig size of 645,701 bp. The M139 genome includes 5,046,600 bp with a GC content of 64%.

We compared the sequenced genome of M139 with the published reference genome of M. abscessus strain ATCC 19977 in order to search for common genomic regions (core regions) and strain-specific regions (accessory regions), using the Pan-genomic Sequence (Panseq) web server (4). Information from these regions is expected to provide insight into species diversity, genetic determinants of basic lifestyle, and strain-specific properties such as environmental survival (8). In our analysis, Panseq identified core regions with a total size of 4,857,651 bp and a GC content of 64% and accessory regions (M139 strain specific) of 481,453 bp with a GC content of 61%. Further annotation using the Rapid Annotation using Subsystems Technology (RAST) pipeline (1) revealed 4,675 coding sequences (CDS) and 43 RNAs in the core genome. There are 419 putative genes distributed in the subsystem of cofactors, vitamins, prosthetic groups, and pigments and 254 genes in the subsystem of carbohydrates. In the accessory genome, there are 525 CDS, including 10 putative genes in the subsystem of phages, prophages, transposable elements, and plasmids which might contribute to species diversity.

To determine the subspecies classification for M139, we performed a phylogenetic analysis first with rpoB and hsp65 genes separately and then with both genes concatenated. Both phylogenetic trees showed M139 clustering with Mycobacterium abscessus subsp. massiliense. This subspecies has been reported to be associated with erm41 (erythromycin ribosome methyltransferase) gene deletions at positions 64 and 65, as well as a 276-bp deletion after position 158 (3). These features have been used to differentiate M. abscessus subsp. massiliense from the other M. abscessus subspecies of M. abscessus sensu stricto and Mycobacterium bolletii. However, we found no deletions at these positions in the erm41 gene in strain M139. Conversely, the erm41 gene sequence of M139 shows the nucleotide variations observed by Kim et al. (3) in the erm41 gene of M. abscessus sensu stricto. These conflicting preliminary genomic observations will be further investigated with the use of other gene targets, and on a larger number of strains.

**Nucleotide sequence accession numbers.** The M. abscessus strain M139 genome sequence and annotation data have been deposited in NCBI GenBank under the accession number AKVR00000000. The version described in this paper is the first version, AKVR01000000.

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