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GENOME ANNOUNCEMENT

Whole-Genome Sequence of the Hypervirulent Clinical Strain *Mycobacterium intracellulare* M.i.198

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We report herein the draft genome sequence of *Mycobacterium intracellulare* clinical strain M.i.198, which consistently exhibits hypervirulence in human patients, human macrophages *in vitro*, and immunocompetent mice.

Nontuberculous mycobacteria are of great clinical importance with respect to the recent increasing prevalence of nontuberculous mycobacterioses, such as *Mycobacterium avium* complex (MAC) disease, which is difficult to treat without specific antibiotics (1). However, the bacteriological etiology of nontuberculous mycobacteria remains to be elucidated. We identified a hypervirulent *Mycobacterium intracellulare* strain (M.i.198) from an immunocompetent patient with pulmonary MAC disease (5). To help understand the genetic background of the virulence, we performed whole-genome sequencing of strain M.i.198.

We sequenced *M. intracellulare* M.i.198 genomic DNA on a Roche 454 FLX Titanium sequencer and assembled the reads using the software program Newbler v2.3. A total of 924,616 reads was generated, with an average read length of 246 bp, yielding a total sequence of 227,524,944 bp.

The assembled sequences contained 149 contigs, and the length of all contigs combined was 5,406,664 bp, with a G+C ratio of 68.0%. The average coverage depth was 42.1×, the N_{50} contig size was 90,025 bp, the average contig was 40,008 bp long, and the longest contig was 453,296 bp. The final assembly comprised 122 contigs in five scaffolds (99.7% of contig bases). We constructed an optical map (OpGen) of strain M.i.198 with the NheI restriction enzyme, yielding 311 ordered restriction fragments (average fragment size, 16 kb). The genome size was estimated to be approximately 5.22 Mb; this was obtained from the sum of all restriction fragments. The five scaffolds were placed on the map, confirming a single circular chromosome without plasmids.

Genome annotation using the NCBI Prokaryotic Genomes Automatic Annotation Pipeline (http://www.ncbi.nlm.nih.gov/genomes/static/Pipeline.html) identified 5,146 coding sequences (CDSs). Strain M.i.198 has three rRNAs (in a single rRNA operon) and 48 tRNA genes.

We compared the sequence of M.i.198 with those of three other recently sequenced *M. intracellulare* strains: ATCC 13950 (GenBank accession no. CP003322.1), MOTT-02 (GenBank accession no. CP003323.1), and MOTT-64 (GenBank accession no. CP003324.1)—the latter two were clinical isolates from Korean patients (2, 3, 4). The reciprocal best-hit BLAST approach revealed that strain M.i.198 shares 90.0%, 92.7%, and 88.7% of its CDSs with *M. intracellulare* ATCC 13950, MOTT-02, and MOTT-64, respectively. Of particular interest is a 51-kb region of difference, 55 CDSs in length (G+C ratio, 61.0%), which consists of prophages in M.i.198. The genome sequence of strain M.i.198 will provide an invaluable resource for understanding the microbiological aspects of human-pathogen interactions, especially the pathogenesis of human MAC disease.

Nucleotide sequence accession numbers. The whole-genome sequence of M.i.198 has been deposited in DDBJ/EMBL/GenBank under the accession numbers BAGQ01000001 to BAGQ01000149.

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REFERENCES

1. **Griffith DE, et al.** 2007. An official ATS/IDSA statement: diagnosis, treatment, and prevention of nontuberculous mycobacterial diseases. Am. J. Respir. Crit. Care Med. **175**:367–416.
2. **Kim BJ, Choi BS, Lim JS, Choi IY, Kook YH.** 2012. Complete genome sequence of *Mycobacterium intracellulare* clinical strain MOTT-64, belonging to the INT1 genotype. J. Bacteriol. **194**:3268.
3. **Kim BJ, et al.** 2012. Complete genome sequence of *Mycobacterium intracellulare* clinical strain MOTT-02. J. Bacteriol. **194**:2771.
4. **Kim BJ, et al.** 2012. Complete genome sequence of *Mycobacterium intracellulare* strain ATCC 13950^T. J. Bacteriol. **194**:2750.
5. **Tateishi Y, et al.** 2009. Virulence of *Mycobacterium avium* complex strains isolated from immunocompetent patients. Microb. Pathog. **46**:6–12.

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