## Journal of Bacteriology

### **Complete Genome Sequence of** Mycobacterium phlei Type Strain **RÍVM601174**

Abdallah M. Abdallah, Mamoon Rashid, Sabir A. Adroub, Marc Arnoux, Shahjahan Ali, Dick van Soolingen, Wilbert **Bitter and Arnab Pain** J. Bacteriol. 2012, 194(12):3284. DOI: 10.1128/JB.00485-12.

Updated information and services can be found at:

http://jb.asm.org/content/194/12/3284

These include:

**REFERENCES** This article cites 13 articles, 2 of which can be accessed free at:

http://jb.asm.org/content/194/12/3284#ref-list-1

**CONTENT ALERTS** Receive: RSS Feeds, eTOCs, free email alerts (when new

articles cite this article), more»

Information about commercial reprint orders: http://journals.asm.org/site/misc/reprints.xhtml To subscribe to to another ASM Journal go to: http://journals.asm.org/site/subscriptions/



# Complete Genome Sequence of *Mycobacterium phlei* Type Strain RIVM601174

Abdallah M. Abdallah, a Mamoon Rashid, Sabir A. Adroub, Marc Arnoux, Shahjahan Ali, Dick van Soolingen, Wilbert Bitter, and Arnab Pain

Pathogen Genomics Laboratory, Computational Bioscience Research Center, King Abdullah University of Science and Technology, Thuwal-Jeddah, Saudi Arabia<sup>a</sup>; Bioscience Core Laboratory, King Abdullah University of Science and Technology, Thuwal-Jeddah, Saudi Arabia<sup>b</sup>; Dutch National Institute for Public Health and the Environment, Bilthoven, The Netherlands<sup>c</sup>; Department of Clinical Microbiology and Department of Pulmonary Diseases, Radboud University Nijmegen Medical Centre, Nijmegen, The Netherlands<sup>d</sup>; and Department of Medical Microbiology and Infection Control, VU University Medical Center, Amsterdam, The Netherlands<sup>e</sup>

*Mycobacterium phlei* is a rapidly growing nontuberculous *Mycobacterium* species that is typically nonpathogenic, with few reported cases of human disease. Here we report the whole genome sequence of *M. phlei* type strain RIVM601174.

ontuberculous mycobacteria (NTM) are ubiquitous organisms and are increasingly recognized as an important cause of infection in immunocompromised and immunocompetent individuals. Mycobacterium phlei is a fast-growing, saprophytic bacterium that is widely distributed in soil and dust and on plants. It has only occasionally been associated with disease in humans with a suppressed immune system (5-7, 9) or in immunocompetent individuals (4, 8, 10). M. phlei was repeatedly isolated from synovial fluid and tissue in an immunocompetent pediatric patient with conjunctivitis, urethritis, and arthritis (1). Most of the existing identification methods for mycobacterial isolates rely on just a single genetic target, and often diverse variants are grouped in one (sub)species (11). Whole-genome sequencing provides the highest resolution on the DNA level and therefore is the most reliable approach for determining the genetic relatedness of mycobacteria. Information on specific genes, especially those of pathogenic mycobacteria, can be used to identify, unequivocally, subgroupings within species associated with clinical relevance. To facilitate a more reliable genetic identification between and within Mycobacterium species, we have characterized the complete genome sequence of M. phlei strain RIVM601174.

The whole-genome sequencing of the M. phlei type strain RIVM601174 genome was performed on the Illumina Hiseq2000 platform using a paired-end read library of read length 100 bp with insert size of 500 bp. The short sequence reads were first processed with Trimmomatic (http://www.usadellab.org /cms/index.php?page=trimmomatic) and FastQC (http://www .bioinformatics.babraham.ac.uk/projects/fastqc/) software before assembling them using the de novo assembler Velvet (12), resulting in 102 contigs with an N<sub>50</sub> of 155,851 bp, comprising in total 5,681,954 bp. The overall GC content of the chromosome was 69.24%, one of the highest among the mycobacteria. The genome annotation was performed using the NCBI Prokaryotic Genomes Automatic Annotation Pipeline (PGAAP). The M. phlei genome was predicted to include 5,435 coding sequences (CDSs), four sets of rRNA operons, and 50 tRNA-encoding genes. It was possible to assign a biological function to 72% (3,969) of the coding sequences.

The RAST server annotation pipeline (2) was used to reveal that *M. phlei* is most closely related to *Mycobacterium* sp. strain MCS among all mycobacteria for which a complete genome sequence is available. The *M. phlei* genome was found to be smaller (5.64 Mb) than the genome of *Mycobacterium* sp. MCS (5.71 Mb)

and to encode fewer genes (5,489 versus 5,698). Interestingly, unlike rapidly growing mycobacteria, our analysis of the *M. phlei* genome showed that this genome encodes a putative mammalian cell entry (MCE) operon, which was previously shown to be conserved only among slow-growing mycobacteria. This virulence factor operon in nonpathogenic *Escherichia coli* confers the ability to invade and survive inside host cells, such as macrophages and HeLa cells (3, 13). Further genomic and functional analyses are needed to investigate this observation.

Nucleotide sequence accession numbers. The results from this whole-genome shotgun project have been deposited with DDBJ/EMBL/GenBank under accession number AJFJ00000000. The version described in this paper is the first version, AJFJ00000000.

#### **ACKNOWLEDGMENTS**

This work was supported by faculty funding to A.P. by King Abdullah University of Science and Technology. M.R. was supported by a SABIC postdoctoral fellowship.

We thank the Dutch National Institute for Public Health and the Environment (RIVM) for providing the strain. We also acknowledge the support of Eveline Weerdenbrug and Roy Ummels at the Department of Medical Microbiology and Infection Control, VU University Medical Centre, Amsterdam, The Netherlands, and Thomas Dan Otto from The Wellcome Trust Sanger Institute, Cambridge, United Kingdom.

#### REFERENCES

- Aguilar JL, Sanchez EE, Carrillo C, Alarcon GS, Silicani A. 1989. Septic arthritis due to Mycobacterium phlei presenting as infantile Reiters syndrome. J. Rheumatol. 16:1377–1378.
- Aziz RK, et al. 2008. The RAST server: rapid annotations using subsystems technology. BMC Genomics 9:75. doi:10.1186/1471-2164-9-75.
- 3. Haile Y, Caugant DA, Bjune G, Wiker HG. 2002. Mycobacterium tuberculosis mammalian cell entry operon (mce) homologs in Mycobacterium other than tuberculosis (MOTT). FEMS Immunol. Med. Microbiol. 33:125–132.
- Hsiao CH, Lin YT, Lai CC, Hsueh PR. 2010. Clinicopathologic characteristics of nontuberculous mycobacterial lung disease in Taiwan. Diagn. Microbiol. Infect. Dis. 68:228–235.

Received 27 March 2012 Accepted 9 April 2012

Address correspondence to Abdallah M. Abdallah, Abdallah @kaust.edu.sa, or Arnab Pain, arnab.pain@kaust.edu.sa.

Copyright © 2012, American Society for Microbiology. All Rights Reserved. doi:10.1128/JB.00485-12

- Karnam S, et al. 2011. Mycobacterium phlei, a previously unreported cause of pacemaker infection: thinking outside the box in cardiac device infections. Cardiol. J. 18:687–690.
- 6. Khatter S, Singh UB, Arora J, Rana T, Seth P. 2008. Mycobacterial infections in human immuno-deficiency virus seropositive patients: role of non-tuberculous mycobacteria. Indian J. Tuberc. 55:28–33.
- Paul E, Devarajan P. 1998. Mycobacterium phlei peritonitis: a rare complication of chronic peritoneal dialysis. Pediatr. Nephrol. 12:67–68.
- Shojaei H, Heidarieh P, Hashemi A, Feizabadi MM, Naser AD. 2011.
  Species identification of neglected nontuberculous mycobacteria in a developing country. Jpn. J. Infect. Dis. 64:265–271.
- Singh S, et al. 2007. Nontuberculous mycobacterial infections in Indian AIDS patients detected by a novel set of ESAT-6 polymerase chain reaction primers. Jpn. J. Infect. Dis. 60:14–18.
- 10. Spiegl PV, Feiner CM. 1994. Mycobacterium phlei infection of the foot: a case report. Foot Ankle Int. 15:680–683.
- van Ingen J, et al. 2009. Mycobacterium noviomagense sp. nov.; clinical relevance evaluated in 17 patients. Int. J. Syst. Evol. Microbiol. 59:845–849.
- 12. **Zerbino DR, Birney** E. 2008. Velvet: algorithms for de novo short read assembly using de Bruijn graphs. Genome Res. 18:821–829.
- Zhang F, Xie JP. 2011. Mammalian cell entry gene family of Mycobacterium tuberculosis. Mol. Cell Biochem. 352:1–10.