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GENOME ANNOUNCEMENT

Complete Genome Sequence of *Mycoplasma pneumoniae* Type 2a Strain 309, Isolated in Japan

Tsuyoshi Kenri,[a] Atsuko Horino,[a] Mari Matsui,[a] Yuko Sasaki,[a] Satowa Suzuki,[a] Mitsuo Narita,[b] Hitomi Ohya,[c] Norio Okazaki,[c] and Keigo Shibayama[a]

Department of Bacteriology II, National Institute of Infectious Diseases, Musashimurayama, Tokyo, Japan[a]; Department of Pediatrics, Sapporo Tokushukai Hospital, Shiroishi-ku, Sapporo, Japan[b]; and Kanagawa Prefectural Institute of Public Health, Chigasaki, Kanagawa, Japan[c]

***Mycoplasma pneumoniae* strain 309, a type 2a (subtype 2 variant) strain of this bacterium, has variations in the P1 protein, which is responsible for attachment of the bacterium to host cells. Here, we report the complete genome sequence of *M. pneumoniae* strain 309 isolated from a pneumonia patient in Japan.**

Mycoplasma pneumoniae is a common pathogen that causes atypical pneumonia and bronchitis in humans, particularly in children and young adults (1, 11). Clinical isolates of this bacterium can be classified into two major groups (subtypes 1 and 2) based on nucleotide sequence variations in the *p1* gene, which encodes an essential factor responsible for cytadherence and pathogenesis (6, 7, 9, 10). Here, we report the complete genome sequence of *M. pneumoniae* strain 309, one of the first-discovered type 2a strains, which was isolated in Hokkaido, Japan, in 1998 (6).

The genome was sequenced using a Roche 454 GS Junior sequencer. A single analysis generated 77.2-Mb sequences (151,617 reads; average length, 509 bp), providing approximately 95-fold genome coverage. Sequences were assembled using GS *de novo* assembler v. 2.5p1; 8 contigs, from 416 to 0.5 kb in size, resulted. We combined these contigs into a circular genome using Sanger sequencing of PCR amplicons derived using primers specific to contig termini. During the genome annotation process, we identified 84 suspected sequencing errors caused by 454 pyrosequencing. These sites were resequenced by Sanger sequencing; 23 pyrosequencing errors were confirmed and corrected.

The complete genome of *M. pneumoniae* strain 309 encompasses 817,176 bp of chromosomal DNA (39.98% GC content), containing 707 predicted coding sequences (CDS), 1 rRNA operon, 36 tRNAs, and 4 noncoding RNA genes. This genome is most similar to those of *M. pneumoniae* M129 and FH (GenBank accession numbers NC000912 and CP002077, respectively), subtype 1 and 2 strains, respectively (2, 8).

A notable difference between these genomes and that of strain 309 is a 6-kb insertion at MPNA5870 in strain 309; this position corresponds to MPN586 of the M129 genome. The M129 and FH genomes are nearly identical at this position. MPNA5870 (MPN586) and several neighboring CDS are putative lipoprotein-encoding genes that are similar to each other but not identical. The 6-kb insertion in the strain 309 genome contains five additional putative lipoprotein genes. Unexpectedly, comparison of the M129, FH, and 309 strains from our laboratory by PCR revealed that strain FH also included an approximately 5-kb insertion at this position. Whether the FH in our laboratory and the genome-sequenced FH strains are in fact different is unclear. However, it is likely that *M. pneumoniae* strains vary in this region, involving a change in the number of putative lipoprotein genes.

Type 2a strains were rarely detected in the 1990s, but, after 2003, they have frequently been found in clinical specimens in Japan (5), consistent with reports from other countries (3, 4, 12). Precise comparisons of strain 309 and other *M. pneumoniae* genomes will identify differences that affect surface molecules, such as lipoproteins or cytadherence proteins, thereby changing their antigenicity. Such information is crucial for understanding the recent increase of type 2a strains. The genome sequence reported here may also be useful in developing strategies for treatment of *M. pneumoniae* infections.

Nucleotide sequence accession number. The sequence data for *M. pneumoniae* strain 309 have been deposited in DDBJ/EMBL/GenBank databases under accession number AP012303.

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Address correspondence to Tsuyoshi Kenri, kenri@nih.go.jp.

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