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GENOME ANNOUNCEMENT

Complete Genome Sequences of *Mycoplasma leachii* Strain PG50^T and the Pathogenic *Mycoplasma mycoides* subsp. *mycoides* Small Colony Biotype Strain Gladysdale

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***Mycoplasma mycoides* subsp. *mycoides* small colony biotype (SC) is the high-consequence animal pathogen causing contagious bovine pleuropneumonia. We report the complete genome sequences of the pathogenic strain *M. mycoides* subsp. *mycoides* SC Gladysdale and a close phylogenetic relative, *Mycoplasma leachii* PG50^T, another bovine pathogen of the *M. mycoides* phylogenetic clade.**

Contagious bovine pleuropneumonia (CBPP) is among the most important blights of cattle in countries in which CBPP is endemic (8). Although the study of the etiologic agent, *Mycoplasma mycoides* subsp. *mycoides* small colony biotype (SC), has benefited greatly from the availability of the genome sequence of PG1^T (9), the avirulence of this reference isolate (2) limits understanding of pathogenic strains. Reported herein is the complete genome sequence of *M. mycoides* subsp. *mycoides* SC strain Gladysdale, a pathogenic isolate (6), which is also employed at the Foreign Animal Disease Diagnostic Laboratory at the USDA Plum Island Animal Disease Center (PIADC) to experimentally demonstrate acute-disease pathology.

Genomic DNA from a clonal isolate (MU clone SC5) was prepared at PIADC and safety tested before transfer to the J. Craig Venter Institute (JCVI; Rockville, MD). The genome was sequenced to 8× coverage by the whole-genome shotgun (WGS) approach using paired-end Sanger sequencing. Scaffold-directed gap closure yielded a single complete sequence. This resulting assembly was autoannotated via the JCVI pipeline, with manual curation applied to specific genes encoding phase-variable surface lipoproteins or associated with genomic islands. The circular chromosome (1,193,808 bp) is slightly smaller than that of *M. mycoides* subsp. *mycoides* SC PG1^T (1,211,703 bp) due primarily to copy number variation of discrete gene blocks. No prophages or prototypic mycoplasmal integrative conjugative element (ICE) units occur, although a region with similarity to *Mycoplasma capricolum* Tra Island I (5) is present in both strains. Gene sets that endow cytotoxic H_2O_2 production, the principal virulence factor identified for *M. mycoides* subsp. *mycoides* SC (4), also occur in both strains. Accordingly, we surmise that differences in virulence may be enciphered not by macroscale insertion or deletion of genomic regions but rather within the multiple single nucleotide polymorphisms (SNPs) and indel differences dispersed throughout the chromosomes.

To enable comparative genomic analyses of bovine pathogens belonging to the *M. mycoides* phylogenetic clade (2), WGS Sanger sequencing (paired-end approach) and assembly were similarly completed for *Mycoplasma leachii* strain PG50^T (MU clone A8). This species, formerly known as *Mycoplasma* sp. bovine group 7 (2), causes pneumonia, mastitis, polyarthritis, and abortion (7). Notable features of the 1,008,951-bp chromosome are the paucity of insertion sequence (IS) incursions in comparison to the IS-laden genomes of *M. mycoides* subsp. *mycoides* SC strains and the presence of an integrative element that encodes multiple palindromic amphipathic repeat coding element (PARCEL) domain proteins (5). In addition, a novel portfolio of phase-variable lipoprotein genes, predicting combinatorial expression governed by indel mutations in poly(TA) tract-containing promoters, verifies the widespread presence of this stochastic mechanism of surface diversification among pathogens of the *M. mycoides* clade (3, 10) and further supports its likely role in host niche adaptation, immune avoidance, or disease transmission.

With genome sequences determined for all taxa within the *M. mycoides* clade (2), a framework is established to expound the pangenome of this group and to identify possible gene patterns that correlate with ruminant host specificity or disease severity. Furthermore, as members of this group are model organisms for genome synthesis and transplantation (1), these data should be a valuable adjunct in designing novel genomes through synthetic biology.

Nucleotide sequence accession numbers. The complete genome sequences are available in GenBank under accession numbers CP002107.1 (*M. mycoides* subsp. *mycoides* SC Gladysdale) and CP002108.1 (*M. leachii* PG50^T).

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