



Draft Genome Sequence of "Candidatus Mycoplasma haemobos," a Hemotropic Mycoplasma Identified in Cattle in Mexico

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We present here the draft genome sequence of the first "*Candidatus* Mycoplasma haemobos" strain found in cattle in Mexico. This hemotropic mycoplasma causes acute and chronic disease in animals. This genome is a starting point for studying the role of this mycoplasma in coinfections and synergistic mechanisms associated with the disease.

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emotropic mycoplasmas (hemoplasmas) are the causative agent of infectious anemia in a variety of mammalian species (1). Bovine hemoplasmas are cell wall-less bacteria that can be found attached to surface-bound or free erythrocytes in the plasma of cattle (2). "Candidatus Mycoplasma haemobos" (syn. "Candidatus Mycoplasma haemobovis) may cause anemia and depression to various degrees, edema, reproductive problems, and various other clinical signs in cattle (3). These bacteria have been reported in cattle from several countries around the world and may act in a synergistic manner when coinfections are present (4-8). In this study, we report the draft genome sequence of "Candidatus Mycoplasma haemobos" INIFAP01, an uncultivated hemoplasma found in blood of sick cattle from Chihuahua, Mexico. Genomic DNA was obtained using the Ultra-Clean DNA BloodSpin kit (Mo Bio Laboratories). Five micrograms of genomic DNA were sequenced in the HiSeq 2000 system (Illumina). We obtained a random data set of 45,165,900 paired-end reads of 300 bases long. Quality-based trimming was performed with a DynamicTrim (SolexaQA++) Perl script, and genome assembly was accomplished de novo using the SPAdes (version 3.1.1) program. The draft genome has 18 contigs, with a calculated 935,638-bp total length, N_{50} contig size of 256,799 bp, G+C content of 30.46% and \sim 48× coverage.

We identified the 16S rRNA gene of "*Ca.* Mycoplasma haemobos" INIFAP01 using the RNAmmer 1.2 server (http://www.cbs .dtu.dk/services/RNAmmer/) and compared it with 30 16S rRNA gene sequences of the genus *Mycoplasma* and two 16S rRNA genes of the genus *Ureaplasma* (outgroup). All sequences were aligned with the MUSCLE server (http://phylogeny.lirmm.fr /phylo_cgi/one_task.cgi?task_type=muscle), and a phylogenetic analysis was performed in the MEGA (version 6.1) program with a neighbor-joining algorithm using 1,000 replicates for bootstrapping. Phylogenetic analysis reveals that "*Ca.* Mycoplasma haemobos" INIFAP01 is closely related to the *Mycoplasma haemobos* species. The 16S rRNA gene of "*Ca.* Mycoplasma haemobos" INIFAP01 is 1,460 bp in length.

The 18 contigs were analyzed with the RAST server (http://rast .nmpdr.org/), identifying 1,216 open reading frames (ORFs) and

1,184 coding sequences (CDSs). This study was approved by the Centro Nacional de Investigación Disciplinaria en Parasitología Veterinaria (CENID-PAVET) branch of the Instituto Nacional de Investigaciones Forestales Agrícolas y Pecuarias (INIFAP) Animal Experimentation and Ethics Committee and conducted considering ethic and methodological aspects, in agreement with the Mexican regulations related to use, housing, and transport of experimental animals, no. NOM-062-ZOO-1999.

Nucleotide sequence accession numbers. This whole-genome shotgun project has been deposited at DDBJ/ENA/GenBank under the accession no. LWUJ000000000. The version described in this paper is version LWUJ01000000.

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We declare no conflicts of interest regarding the publication of this paper.

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REFERENCES

- Willi B, Boretti FS, Tasker S, Meli ML, Wengi N, Reusch CE, Lutz H, Hofmann-Lehmann R. 2007. From haemobartonella to hemoplasma: molecular methods provide new insights. Vet Microbiol 125:197–209. http:// dx.doi.org/10.1016/j.vetmic.2007.06.027.
- Girotto A, Zangirólamo AF, Bogado ALG, Souza ASLE, da Silva GCF, Garcia JL, Vilas Boas LA, Biondo AW, Vidotto O. 2012. Molecular detection and occurrence of "*Candidatus* Mycoplasma haemobos" in dairy cattle of southern Brazil. Rev Bras Parasitol Vet 21:342–344. http:// dx.doi.org/10.1590/S1984-29612012000300034.
- 3. Hornok S, Micsutka A, Meli ML, Lutz H, Hofmann-Lehmann R. 2011.

Molecular investigation of transplacental and vector-borne transmission of bovine haemoplasmas. Vet Microbiol 152:411–414.

- Meli ML, Willi B, Dreher UM, Cattori V, Knubben-Schweizer G, Nuss K, Braun U, Lutz H, Hofmann-Lehmann R. 2010. Identification, molecular characterization, and occurrence of two bovine hemoplasma species in Swiss cattle and development of real-time TaqMan quantitative PCR assays for diagnosis of bovine hemoplasma infections. J Clin Microbiol 48: 3563–3568. http://dx.doi.org/10.1128/JCM.02224-09.
- Tagawa M, Matsumoto K, Inokuma H. 2008. Molecular detection of *Mycoplasma wenyonii* and "*Candidatus* Mycoplasma haemobos" in cattle in Hokkaido, Japan. Vet Microbiol 132:177–180. http://dx.doi.org/10.1016/ j.vetmic.2008.05.006.
- 6. Gladden N, Haining H, Henderson L, Marchesi F, Graham L, McDonald

M, Murdoch FR, Bruguera Sala A, Orr J, Ellis K. 2015. A case report of *Mycoplasma wenyonii* associated immune-mediated haemolytic anaemia in a dairy cow. Ir Vet J 69:1. http://dx.doi.org/10.1186/s13620-016-0061-x.

- De la Fuente J, Ruybal P, Mtshali MS, Naranjo V, Shuqing L, Mangold AJ, Rodríguez SD, Jiménez R, Vicente J, Moretta R, Torina A, Almazán C, Mbati PM, de Echaide ST, Farber M, Rosario-Cruz R, Gortazar C, Kocan KM. 2007. Analysis of world strains of *Anaplasma marginale* using major surface protein 1a repeat sequences. Vet Microbiol 119:382–390. http://dx.doi.org/10.1016/j.vetmic.2006.09.015.
- Su QL, Song HQ, Lin RQ, Yuan ZG, Yang JF, Zhao GH, Huang WY, Zhu XQ. 2010. The detection of "*Candidatus* Mycoplasma haemobos" in cattle and buffalo in China. Trop Anim Health Prod 42:1805–1808. http:// dx.doi.org/10.1007/s11250-010-9640-0.