

Whole-Genome Sequence of *Mesorhizobium hungaricum* sp. nov. Strain UASWS1009, a Potential Resource for Agricultural and Environmental Uses

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We report here the whole-genome shotgun sequences of the strain UASWS1009 of the species *Mesorhizobium hungaricum* sp. nov., which are different from any other known *Mesorhizobium* species. This is the first genome registered for this new species, which could be considered as a potential resource for agriculture and environmental uses.

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The genus *Mesorhizobium*, established in 1997, gathers rhizobacterial species genetically different from *Rhizobium* species (1). *Mesorhizobium* bacteria are mobile, aerobic, Gram-negative, and non-spore-forming rods and have a G+C content between 59 and 64% (2). These nitrogen-fixing bacteria may usually be found in symbiotic nodules or as endophytes in mimosoid temperate legumes (2). Strain UASWS1009 was isolated from the sewage sludge of a coking plant in Hungary through a selection experiment for highly ammonia-tolerant nitrifying bacteria. Initially identified as a *Mesorhizobium* species by 16S sequencing, it shared 98 to 99% identity with many *Mesorhizobium* strains in GenBank (3). Genomic DNA was extracted (4) and fragmented to an average size of 350 bp in a 50- μ l Adaptive Focused Acoustics (AFA) microTUBE (Covaris, USA) in an S2 ultrasonicator (Covaris). The TruSeq DNA PCR-free library preparation kits (Illumina, USA) produced a library, of which insert sizes were checked in a fragment analyzer (Advanced Analytical Technologies, Inc.). Whole-genome shotgun (WGS) sequencing was performed in one Illumina MiniSeq run at 2 \times 150-bp paired-end read length, using a MiniSeq Mid output kit (Illumina). The sequencing yielded 4,538,758 reads (680 Mb of DNA) providing 108-fold genome coverage. The reads are available from the NCBI Sequence Read Archive (SRA) database under the accession no. SRR4031082. Following quality control with FastQC (5) and assembly with SPAdes genome assembler 3.8.1 (6), the contigs were arranged with BioEdit (7) and analyzed with QUAST (8). The final assembly produced 41 contigs (\geq 200 bp), with a total genome length of 6,303,257 bp, a G+C content of 63.02%, and an N_{50} value of 631,149 bp. No plasmid was found by PlasmidFinder (9) and plasmidSPAdes (10). While RAST version 2.0 (11) annotated 6,111 coding sequence (CDS) genes distributed in 484 subsystems, the Prokaryotic Genome Automatic Annotation Pipeline (PGAAP) (12) identified 5,972 genes for 5,916 CDSs and 5,849 coding genes, 67 pseudogenes, and 56 RNA genes. A Mu prophage genome (30 genes) is integrated in contig 41. Mobile elements (22

sequences) are concentrated on contigs 22, 26, and 32. No virulence, disease, or toxin genes were found. It is equipped with genes for antibiotic production and resistance genes against metals (arsenic, cadmium, chrome, cobalt, copper, mercury, and zinc) and against a few antibiotics (penicillin, fluoroquinolones, streptomycin, and clavulanic acid). Diverse degradation pathways of aromatic compounds are provided by 80 genes, offering a potential use for environmental treatment of contaminated soils and water. Plant growth promotion properties are provided by genes for siderophore synthesis and transport (12 genes), 1-aminocyclopropane-1-carboxylate deaminase (ACC) for ethylene degradation (one gene), genes for plant hormones (13 genes), and antimicrobial compounds. Specific to plant-microbe associations, a large type IV secretion system of 35 genes was identified (13). The sequence similarity was found to be less than 80% with its closest phylogenetic neighbor *Mesorhizobium amorphae* strain CCN-WGS0123 (14), which has a much larger genome. Contrary to this species, strain UASWS1009 had no *nif* and *nod* genes. As an environmental isolate, its legume hosts spectrum remains unknown. We propose the name *Mesorhizobium hungaricum* sp. nov. Crovadore and Lefort due to its geographical origin.

Accession number(s). This whole-genome shotgun (WGS) project was deposited at DDBJ/EMBL/GenBank under the accession no. [MDEO00000000](https://www.ncbi.nlm.nih.gov/nuccore/MDEO00000000). The version described in this paper is the first version, MDEO00000000.1. The 41 contigs have been deposited under the accession numbers MDEO01000001 to MDEO01000041.

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REFERENCES

- Jarvis BDW, Van Berkum P, Chen WX, Nour SM, Fernandez MP, Cleyet-Marel JC, Gillis M. 1997. Transfer of *Rhizobium loti*, *Rhizobium*

- huakuui*, *Rhizobium ciceri*, *Rhizobium mediterraneum*, and *Rhizobium tianshanense* to *Mesorhizobium* gen. nov. *Int J Syst Bacteriol* 47:895–898. <http://dx.doi.org/10.1099/00207713-47-3-895>.
2. Laranjo M, Alexandre A, Oliveira S. 2014. Legume growth-promoting rhizobia: an overview on the *Mesorhizobium* genus. *Microbiol Res* 169: 2–17. <http://dx.doi.org/10.1016/j.micres.2013.09.012>.
 3. Benson DA, Cavanaugh M, Clark K, Karsch-Mizrachi I, Lipman DJ, Ostell J, Sayers EW. 2013. GenBank. *Nucl Acids Res* 41:D36–D42. <http://dx.doi.org/10.1093/nar/gks1195>.
 4. Lefort F, Douglas GC. 1999. An efficient micro-method of DNA isolation from mature leaves of four hardwood tree species *Acer*, *Fraxinus*, *Prunus* and *Quercus*. *Ann For Sci* 56:259–263. <http://dx.doi.org/10.1051/forest:19990308>.
 5. Andrews S. 2010. FastQC: a quality control tool for high throughput sequence data. <http://www.bioinformatics.babraham.ac.uk/projects/fastqc>.
 6. Bankevich A, Nurk S, Antipov D, Gurevich AA, Dvorkin M, Kulikov AS, Lesin VM, Nikolenko SI, Pham S, Prjibelski AD, Pyshkin AV, Sirotkin AV, Vyahhi N, Tesler G, Alekseyev MA, Pevzner PA. 2012. SPAdes: a new genome assembly algorithm and its applications to single-cell sequencing. *J Comput Biol* 19:455–477. <http://dx.doi.org/10.1089/cmb.2012.0021>.
 7. Hall TA. 1999. BioEdit: a user-friendly biological sequence alignment editor and analysis program for Windows 95/98/NT, *Nucleus Acids Symp Ser* 41:95–98.
 8. Gurevich A, Saveliev V, Vyahhi N, Tesler G. 2013. QUAST: quality assessment tool for genome assemblies. *Bioinformatics* 29:1072–1075. <http://dx.doi.org/10.1093/bioinformatics/btt086>.
 9. Carattoli A, Zankari E, García-Fernández A, Voldby Larsen M, Lund O, Villa L, Møller Aarestrup F, Hasman H. 2014. *In silico* detection and typing of plasmids using PlasmidFinder and plasmid multilocus sequence typing. *Antimicrob Agents Chemother* 58:3895–3903. <http://dx.doi.org/10.1128/AAC.02412-14>.
 10. Antipov D, Hartwick N, Shen M, Raiko M, Lapidus A, Pevzner P. July 27, 2016. plasmidSPAdes: assembling plasmids from whole genome sequencing data. *Bioinformatics* [Epub ahead of print.] <http://dx.doi.org/10.1093/bioinformatics/btw493>.
 11. Overbeek R, Olson R, Pusch GD, Olsen GJ, Davis JJ, Disz T, Edwards RA, Gerdes S, Parrello B, Shukla M, Vonstein V, Wattam AR, Xia F, Stevens R. 2014. The SEED and the rapid annotation of microbial genomes using subsystems technology (RAST). *Nucl Acid Res* 42: D206–D214. <http://dx.doi.org/10.1093/nar/gkt1226>.
 12. Tatusova T, DiCuccio M, Badretdin A, Chetvernin V, Ciufu S, Li W. 2013. Prokaryotic genome annotation pipeline. *NCBI Handbook*, 2nd ed. National Center for Biotechnology Information, Bethesda, MD.
 13. Schmeisser C, Liesegang H, Krysciak D, Bakkou N, Le Quéré A, Wollherr A, Heinemeyer I, Morgenstern B, Pommerening-Röser A, Flores M, Palacios R, Brenner S, Gottschalk G, Schmitz RA, Broughton WJ, Perret X, Strittmatter AW, Streit WR. 2009. *Rhizobium* sp. strain NGR234 possesses a remarkable number of secretion systems. *Appl Environ Microbiol* 75:4035–4045. <http://dx.doi.org/10.1128/AEM.00515-09>.
 14. Hao X, Lin Y, Johnstone L, Baltrus DA, Miller SJ, Wei G, Rensing C. 2012. Draft genome sequence of plant growth-promoting rhizobium *Mesorhizobium amorphae*, isolated from zinc-lead mine tailings. *J Bacteriol* 194:736–737. <http://dx.doi.org/10.1128/JB.06475-11>.