

# Draft Genome Sequence of *Mesorhizobium alhagi* CCNWXJ12-2<sup>T</sup>, a Novel Salt-Resistant Species Isolated from the Desert of Northwestern China

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***Mesorhizobium alhagi* strain CCNWXJ12-2<sup>T</sup> is a novel species of soil-dwelling, nitrogen-fixing bacteria that can form symbiotic root nodules with *Alhagi sparsifolia*. Moreover, the strain has high resistance to salt and alkali. Here we report the draft genome sequence of *Mesorhizobium alhagi* strain CCNWXJ12-2<sup>T</sup>. A large number of osmotic regulation-related genes have been identified.**

Rhizobia are well known for their ability to establish nitrogen-fixing symbiotic relationships with leguminous plants by forming root nodules (4, 6). *Mesorhizobium alhagi* strain CCNWXJ12-2<sup>T</sup> is the wild microsymbiont of *Alhagi sparsifolia*, which is a pioneer plant species in windbreak and sand fixation in northwest China (13, 14). It has been proven that *Mesorhizobium alhagi* strain CCNWXJ12-2<sup>T</sup> shows high resistance to salt (0.8 Mol/liter NaCl) and alkali (pH 12) (unpublished data). In addition, it has been identified as a novel *Mesorhizobium* species, with the highest 16S rRNA gene sequence similarity being less than 97.8% (2). Here, we present a draft genome sequence of the CCNWXJ12-2<sup>T</sup> type strain.

The genome of *Mesorhizobium alhagi* strain CCNWXJ12-2<sup>T</sup> was sequenced on the Illumina HiSeq 2000 platform by BGI-Shenzhen, China. The genome was assembled using SOAP *de novo* (<http://soap.genomics.org.cn/>). Before assembly, a series of filtering steps were undertaken to filter the low-quality sequencing reads. The retained reads were used to build the contigs, and then all the paired-end reads were realigned onto the contig sequences to construct the scaffolds. A total of 42 scaffolds containing 398 contigs were generated, with 62.65% average GC content. The genome size (7,081,202 nucleotides [nt]) is smaller than that of *Mesorhizobium loti* MAFF303099 (7,596,297 nt) but larger than those of *Mesorhizobium* sp. strain BNC1 (4,935,185 nt), *Mesorhizobium opportunistum* WSM2075 (6,854,796 nt) and *Mesorhizobium ciceri* biovar *biserrulae* WSM1271 (6,690,028 nt) (<http://www.ncbi.nlm.nih.gov/genome/>).

The *de novo* gene prediction was carried out using Glimmer 3.0 software (3). A total of 7,408 open reading frames (ORFs) were predicted. The gene coding sequences covered 86.17% of the genome, and the average length of the coding sequences was 823 bp. Among all the predicted genes, 1,034 genes were expected to code hypothetical or unknown proteins according to BLAST comparisons against several protein sequences and protein family databases such as KEGG, COG, Swiss-Prot, TrEMBL, and NR (1, 9, 12). RNAmmer (8) and tRNAscan-SE (11) were utilized to predict rRNAs and tRNAs. 5S, 16S, and 23S rRNAs (at least one copy each) were identified in the genome. In addition, 46 tRNAs and 2 small RNAs (sRNAs) were found by *de novo* prediction. The NCBI Prokaryotic Genomes Automatic Annotation Pipeline (PGAAP) was employed for gene annotation in preparation for submission

of the sequence information to GenBank (<http://www.ncbi.nlm.nih.gov/genomes/static/Pipeline.html>).

The *Mesorhizobium alhagi* strain CCNWXJ12-2<sup>T</sup> genome carries multiple gene systems potentially involved in salt resistance that allow the cells to respond and adapt to salinity stress. The metabolic pathway analysis of the osmotic regulation network performed using the KEGG databases showed that 377 genes were involved in osmoregulation, including abundant genes encoding members of various transportation system families, Na<sup>+</sup>:H<sup>+</sup> antiporter NhaA families, monovalent cation:H<sup>+</sup> antiporter CPA2 families, two-component system OmpR, NtrC, and LuxR families, glycine betaine/proline transport system ProW, ProV, and ProX families, and potassium transport-related KefB, TrkA, and TrkH systems (5, 7, 10). Nearly 1,200 genes were seen to be involved in environmental information processing-membrane transport. Families corresponding to carbohydrate metabolism, amino acid metabolism, and xenobiotics biodegradation metabolism have the largest number of gene members, accounting for 30% of the genome. Moreover, zinc/manganese transport system, peptide/nickel transport system, and Cu<sup>2+</sup>-exporting ATPase family members were also found.

**Nucleotide sequence accession numbers.** The draft genome sequence has been deposited in GenBank under accession number AHAM00000000. The version described in this paper is the first version, AHAM01000000.

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## REFERENCES

1. Boeckmann B, et al. 2003. The SWISS-PROT protein knowledgebase and its supplement TrEMBL in 2003. *Nucleic Acids Res.* 31:365–370.
2. Chen WM, Zhu WF, Bontemps C, Young JP, Wei GH. 2010. *Mesorhi-*

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- zobium alhagi sp nov., isolated from wild *Alhagi sparsifolia* in north-western China. *Int. J. Syst. Evol. Microbiol.* **60**(Pt. 4):958–962.
3. Delcher AL, Bratke KA, Powers EC, Salzberg SL. 2007. Identifying bacterial genes and endosymbiont DNA with Glimmer. *Bioinformatics* **23**:673–679.
  4. Elsheikh EAE, Wood M. 1995. Nodulation and N<sub>2</sub> fixation by soybean inoculated with salt-tolerant rhizobia or salt-sensitive bradyrhizobia in saline soil. *Soil Biol. Biochem.* **27**:657–661.
  5. Epstein W. 2003. The roles and regulation of potassium in bacteria. *Prog. Nucleic Acid Res. Mol. Biol.* **75**:293–320.
  6. Gage DJ. 2004. Infection and invasion of roots by symbiotic, nitrogen-fixing rhizobia during nodulation of temperate legumes. *Microbiol. Mol. Biol. Rev.* **68**:280–300.
  7. Kashyap DR, Botero LM, Lehr C, Hassett DJ, McDermott TR. 2006. A Na<sup>+</sup>:H<sup>+</sup> antiporter and a molybdate transporter are essential for arsenite oxidation in *Agrobacterium tumefaciens*. *J. Bacteriol.* **188**:1577–1584.
  8. Lagesen K, et al. 2007. RNAmmer: consistent and rapid annotation of ribosomal RNA genes. *Nucleic Acids Res.* **35**:3100–3108.
  9. Ogata H, et al. 1999. KEGG: Kyoto encyclopedia of genes and genomes. *Nucleic Acids Res.* **27**:29–34.
  10. Polarek JW, Williams G, Epstein W. 1992. The products of the Kdpde operon are required for expression of the Kdp atpase of *Escherichia coli*. *J. Bacteriol.* **174**:2145–2151.
  11. Schattner P, Brooks AN, Lowe TM. 2005. The tRNAscan-SE, snoscan and snoGPS web servers for the detection of tRNAs and snoRNAs. *Nucleic Acids Res.* **33**:W686–W689.
  12. Tatusov RL, Galperin MY, Natale DA, Koonin EV. 2000. The COG database: a tool for genome-scale analysis of protein functions and evolution. *Nucleic Acids Res.* **28**:33–36.
  13. Wei GH, Chen WM, Young JPW, Bontemps C. 2009. A new clade of Mesorhizobium nodulating *Alhagi sparsifolia*. *Syst. Appl. Microbiol.* **32**: 8–16.
  14. Zeng FJ, et al. 2002. Water relation characteristics of *Alhagi sparsifolia* and consequences for a sustainable management. *Sci. China Ser. D* **45**: 125–131.