

Genome sequence of a native nitrogen-fixing *Bradyrhizobium* sp. strain RCAM1614 isolated from lupine nodules

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ABSTRACT This report describes the genome sequence of *Bradyrhizobium* sp. strain RCAM1614, which was isolated from the root nodules of *Lupinus angustifolius* L. in the Leningrad district of Russia. The genome has a size of 9.77 Mbp and shows 96.1% completeness, which enables precise taxonomic classification.

KEYWORDS legume-rhizobial symbiosis, symbiotic nodules, *Bradyrhizobium barranii*

Species of the genus *Bradyrhizobium* are recognized as important nitrogen-fixing symbionts of various legumes, including lupine, groundnut, and soybean (1–3). Strain RCAM1614 was isolated according to Vincent (4) approximately 20 years ago from effective nodules of *Lupinus angustifolius* L. in the Leningrad district, Russia and deposited into the network collection of bioresources in the field of genetic technologies for agriculture (RCAM) as *Bradyrhizobium* sp.

Strain was preserved in yeast extract mannitol (YEM) (5) medium with 40% glycerol at –80°C. For DNA isolation, strain was cultivated on YEM at 28°C for 5 days. Genomic DNA was extracted using the Monarch Genomic DNA Purification Kit T3010L (New England Biolabs, USA) according to the manufacturer's recommendations without size selection.

Oxford Nanopore Technologies (ONT) sequencing was carried out in the All-Russia Research Institute for Agricultural Microbiology (ARRIAM). Sequencing libraries were prepared using the Native Barcoding Kit 24 V14 (SQK-NBD114.24) (Oxford Nanopore, United Kingdom) according to the manufacturer's instructions, skipping the DNA-shearing step, and sequenced on a MinION (Oxford Nanopore) device with FLO-MIN114 flow cell. Basecalling was performed with Dorado v1.0.1 (<https://github.com/nanopore-tech/dorado/>) using the dna_r10.4.1_e8.2_400bps_sup@v5.2.0 model. Raw read quality control was performed with NanoPlot (6) v1.44.1. In total, 285,977 reads with N₅₀ = 6,796 bp were obtained.

The genome was assembled *de novo* using Flye (7) v2.9.6 with the (--nano-rw) option, followed by polishing with Medaka v1.7.2 (<https://github.com/nanopore-tech/medaka>). Assembly quality was assessed with QUAST (8) v5.3.0 and completeness was evaluated with BUSCO (9, 10) v5.8.0 using the rhizobiales_odb10 data set. The genome assembly was deposited at GenBank and annotated using the NCBI Prokaryotic Genome Annotation Pipeline (11) v6.10. Default parameters were used for all listed software unless otherwise noted.

The draft genome consists of 5 contigs, with a total length of 9,773,268 bp, a GC content of 63.48%, an assembly N₅₀ value of 9,330,649 bp, an estimated coverage of 112x, and a completeness of 96.1%. It contains 9,089 protein-coding genes, 55 tRNAs, and 6 rRNAs.

Primary taxonomic identification was performed using the Type (Strain) Genome Server (TYGS) (12, 13) v401 platform. Phylogenomic analysis based on whole-genome sequences using the Genome BLAST Distance Phylogeny (GBDP) method placed strain RCAM1614 within a well-supported clade alongside *Bradyrhizobium barranii* strains. The

Editor André O. Hudson, Rochester Institute of Technology, Rochester, New York, USA

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The authors declare no conflict of interest.

See the funding table on p. 3.

Received 16 September 2025

Accepted 15 October 2025

Published 4 November 2025

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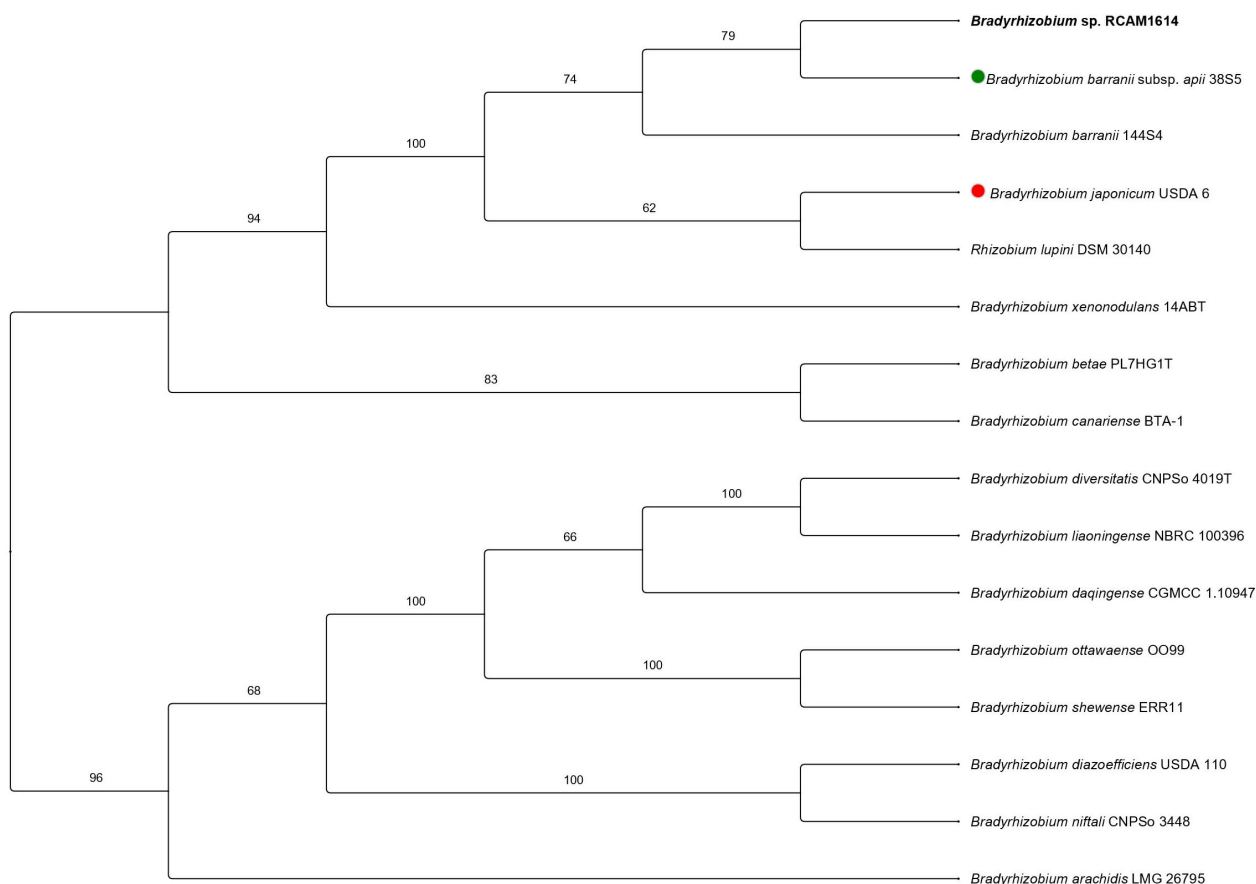


FIG 1 *Bradyrhizobium* sp. RCAM1614 whole-genome based phylogenetic analysis. The numbers above the branches are the GBDP pseudo-bootstrap support values greater than 60%. Strain RCAM1614 (bold), *B. barranii* subsp. *apii* 38S5^T (green dot), *B. japonicum* USDA 6^T (red dot).

most closely related type strain was *B. barranii* subsp. *apii* 38S5^T with a digital DNA-DNA hybridization value of 75.1% (Fig. 1). The final taxonomy check was provided by GenBank with *B. barranii* 97.36% average nucleotide identity match.

ACKNOWLEDGMENTS

This work was supported by the Ministry of Science and Higher Education of the Russian Federation within the framework of the Agreement of May 29, 2025 No. 075-15-2025-472 on the provision of a grant in the form of a subsidy from the federal budget for the implementation of the project: "Expansion of the fund and development of genomic research in the collection of microorganisms the Network Collection of Bioresources in the field of genetic technologies for agriculture (RCAM)".

The research was performed using equipment of the Core Centrum "Genomic Technologies, Proteomics and Cell Biology" in ARRIAM.

Data analysis was performed using the Galaxy server (<https://usegalaxy.org/>).

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FUNDING

Funder	Grant(s)	Author(s)
Ministry of Science and Higher Education of the Russian Federation	Agreement of May 29 2025 No. 075-15-2025-472	Evgenii A. Kirichek Viktor E. Tsyganov

AUTHOR CONTRIBUTIONS

Eugenia Yu. Denisova, Data curation, Investigation, Writing – original draft | Evgenii A. Kirichek, Data curation, Formal analysis, Investigation, Methodology, Supervision, Writing – review and editing | Viktor E. Tsyganov, Conceptualization, Project administration, Supervision, Writing – review and editing

DATA AVAILABILITY

Assembly and sequence data have been deposited in the GenBank under the BioProject accession number [PRJNA1311437](https://ncbi.nlm.nih.gov/bioproject/PRJNA1311437). Assembly has been deposited under the accession number [GCA_052402455.1](https://ncbi.nlm.nih.gov/submit/seq/SRA/GCA_052402455.1). The demultiplexed FASTQ file, with barcodes removed, from the MinION run has been deposited in the Sequence Read Archive (SRA) under number [SRX30289055](https://ncbi.nlm.nih.gov/sra/SRX30289055). This announcement describes the first version of the genome assembly.

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