Arthrobacter rhizophilus sp. nov., an organic phosphate solubilizing bacterium isolated from rhizosphere soil of Meconopsis integrifolia

Yongxia Wang
College of Resources and Environment, Xizang Agricultural and Animal Husbandry University
Chengxin Zhang
Hebei University
Yan Wang
College of Resources and Environment, Xizang Agricultural and Animal Husbandry University
Delong Kong
College of Life Science, Xinjiang Normal University
Zhen Xing
College of Resources and Environment, Xizang Agricultural and Animal Husbandry University
Zhiyong Ruan
ruanzhiyong@caas.cn

Chinese Academy of Agricultural Sciences

Short Report

Keywords: Meconopsis integrifolia, Arthrobacter, rhizosphere soil, organic phosphorus solubilization, polyphasic taxonomy

Posted Date: December 12th, 2025

DOI: https://doi.org/10.21203/rs.3.rs-8214307/v1

Additional Declarations: No competing interests reported.

Abstract

A Gram-staining-positive, rod-shaped, non-motile bacterium, designated strain MI7-26^T, with organic phosphate solubilizing ability was isolated from rhizosphere soil of *Meconopsis integrifolia*, Nyingchi, Xizang, China. Optimal growth conditions were observed at 30 ℃, pH 7.0 and 0.5% (w/v) NaCl. A comparative analysis of 16S rRNA gene sequences revealed that strain MI7-26^T showed the highest similarity to *Arthrobacter bambusae* GM18^T (98.6% sequence similarity); *Arthrobacter gyeryongensis* DCY72^T (98.5%); *Pseudarthrobacter enclensis* NIO-1008^T (97.5%). The genome size was 4.81 Mb and the G + C content was 63.1 mol%. Whole genome-based analysis revealed that MI7-26^T is more closely related to *Arthrobacter ramosus*, *Arthrobacter gyeryongensis*, and *Arthrobacter bambusae*, with both the Average Nucleotide Identity (ANI) and digital DNA-DNA hybridization (dDDH) values below the thresholds used for species delineation (ANI < 90%, dDDH < 40%). The major fatty acids were anteiso-$C_{15:0}$ and anteiso-$C_{17:0}$. The predominant polar lipids were diphosphatidylglycerol, phosphatidylglycerol, phosphatidylinositol, two unidentified phospholipids, one unidentified aminolipid and two unidentified glycolipids. The major quinone was menaquinone MK-9(H_2). The cell-wall peptidoglycan was A3*α* type, containing lysine and alanine. Phenotypically, strain MI7-26^T differed from its close relatives by being positive for the Voges-Proskauer reaction, methyl red test, and hydrolysis of Tween 80, as well as for enzyme activities of β-glucuronidase and N-acetyl-β-glucosaminidase. On the basis of phenotypic, phylogenetic and genotypic data, strain MI7-26^T is considered to represent a novel species of the genus *Arthrobacter*, for which the name *Arthrobacter rhizophilus* sp. nov. is proposed. The type strain is MI7-26^T (= GDMCC 1.3254^T = JCM 35693^T).

INTRODUCTION

Meconopsis integrifolia is a herb of the poppy family that grows in the high mountains of 3400–5000 m [1]. Due to its particularity of its habitat, it has become an endangered species, especially as a Tibetan medicine resource that can treat pneumonia, liver pain, headache and edema [2], which should be paid more attention and protected. Rhizosphere microorganisms were said to be the second genome of plants, especially beneficial microorganisms such as phosphorus-solubilizing bacteria have been reported to promote plant growth through their own secretion of products, or have antagonistic effects on pathogenic bacteria [3], but bacteria in the rhizosphere of *Meconopsis* have rarely been reported.

The genus *Arthrobacter* was first proposed by Conn and Dimmick [4] in 1947, belonging to the family *Micrococcaceae*. According to the List of Prokaryotic Names with Standing in Nomenclature (LPSN; https://lpsn.dsmz.de/genus/Arthrobacter), and currently comprises 74 species of with a validly published and correct name (accessed on 27 October 2025). The genus *Arthrobacter* is widely distributed across diverse environmental sources [5]. Members of the genus *Arthrobacter* are Gram-stain-positive, aerobic, some exhibit a rod-coccus morphological cycle, which contained 55–72 mol% DNA G + C content, and anteiso-$C_{15:0}$ and anteiso-$C_{17:0}$ as the major cellular fatty acids [6]. The quinone system is composed of MK-9(H_2) or MK-8(H_2) as the predominant quinone and A3*α* or A4*α* as the cell-

wall peptidoglycan structural type [7, 8]. In this study, we report a novel species of the genus *Arthrobacter*, an organic phosphate solubilizing bacterium isolated from rhizosphere soil of *M.integrifolia*, designated as MI7-26^T, and characterized using a polyphasic approach.

MATERIALS AND METHODS

ISOLATION, PURIFICATION AND CULTURE OF STRAIN

Strain MI7-26^T was collected from rhizosphere soil of *M.integrifolia*, Nyingchi in Xizang, China (N 29°36'34", E 94°39'17"). One gram of rhizosphere soil was mixed with 9 ml sterile water and shaken at 150 rpm for 30 min. The suspension was serially diluted cultured on the nutrient agar (NA; Difco) medium after at 30 ℃ for 3 days, separated colonies were picked and serially streaked onto NA plates incubating at 30 ℃ to obtain single colony. Strain MI7-26^T was obtained after several re-streaking and transfer onto NA plates. The pure culture of strain MI7-26^T was preserved at -80 ℃ in nutrient broth (NB; Difco) medium with 60% (v/v) glycerol.

PHYLOGENETIC ANALYSIS AND 16S rRNA GENE SEQUENCING

The nearly complete 16S rRNA gene sequence of strain MI7-26T, amplified with primers 27F and 1492R, was compared with sequences of closely related type strains obtained from the EzBioCloud server (https://www.ezbiocloud.net/) [9] using the CLUSTAL X program [10]. Phylogenetic trees were analysed with neighbour-joining (NJ) [11], maximum-likelihood (ML) and maximum-parsimony (MP) [12] by using MEGA 7.0 [13]. Tree topologies were evaluated by bootstrap analysis with 1000 replicates. Evolutionary distances were calculated according to the algorithm of the Kimura's two-parameter model for the NJ, MP and ML trees method. Furthermore, genomic phylogenetic relationships were computed using the Type Genome Server (TYGS; https://tygs.dsmz.de/) [14] and the AutoMLST pipeline (https://automlst.ziemertlab.com) [15], which are based on genome-to-genome distances and concatenated core genes, respectively.

GENOME SEQUENCE ANALYSIS

The genomes of strain MI7-26^T were sequenced on the Illumina MiSeq platform by Guangzhou Magigene Company. Raw reads were *de novo*-assembled using SPAdes (version 3.5.0) [16]. The genomic G + C content was determined by the draft genome sequencing. The digital DNA-DNA hybridization (dDDH) and average nucleotide identity (ANI) were calculated according to the minimal standards proposed by Chun et al. [17]. The dDDH values were calculated by Genome-to-Genome Distance Calculator 3.0 online (http://ggdc.dsmz.de) [18]. The ANI value between two genomes was calculated using the OrthoANIu algorithm (https://www.ezbiocloud.net/tools/ani) [19]. The average amino acid Identity (AAI) was calculated using the Majorbio Cloud Platform (https://analysis.majorbio.com/tools/) [20].

MORPHOLOGICAL, PHYSIOLOGICAL AND BIOCHEMICAL CHARACTERISTICS

The morphological characteristics of strain MI7-26^T were observed after growth on NA medium for 3 days at 30 ℃. The cells were examined by light microscope (Nikon 80i, Tokyo, Japan) and transmission electron microscope (Hitachi 7500, Tokyo, Japan) [21]. The temperature range for growth was determined on NA medium for 7 days at 4, 10, 15, 20, 25, 30, 37, 40 and 45 ℃. NaCl tolerance was checked with different concentrations of NaCl (0, 0.5, 1.0, 2.0, 3.0, 4.0, 5.0, 6.0, 7.0 and 8.0 (%w/v)) added to NB medium (present configuration without sodium chloride). pH dependent growth was evaluated also in NB medium using sterile solutions of citric acid/Na_2HPO_4 (pH 4.0 to 5.0), Na_2HPO_4/NaH_2PO_4 buffer (pH 6.0 to 8.0), $NaHCO_3$/Na_2CO_3 buffer (pH 9.0 to 10.0) or Na_2HPO_4/NaOH buffer (pH 11.0) or KCl/NaOH buffer (pH 12.0). Cell motility was tested by hanging-drop method throughout a tube containing semisolid medium [22]. Gram-stain reaction, catalase and oxidase activities, Voges–Proskauer reaction, H_2S production, hydrolysis of starch, gelatin, casein, cellulose, tyrosine and Tweens 20, 40, 60 and 80 were determined as described by Smibert & Krieg [23]. Additional physiological and biochemical features were determined using the API ZYM, 20NE and 50CH strips (bioMérieux) as described by the manufacturer's instructions.

CHEMOTAXONOMIC ANALYSIS

Strain MI7-26^T and the reference strain were cultured on trypticase soy agar (TSA; Difco) medium at 30 ℃ and cells were harvested in the late exponential phase. Cellular fatty acids analysis was carried out as described by Sakamoto et al. [24]. The identification and quantification of the fatty acid methyl esters as well as the numerical analysis of the fatty acid profiles were carried out by the Sherlock Microbial Identification System with the standard MIS Library Generation Software (VERSION 6.0 and Date 4, Microbial ID) and a 6890N gas chromatograph (Agilent) according to the manufacturers' instructions. Quantitative analysis of the peptidoglycan amino acids was performed by gas chromatography according to the method of MacKenzie et al. [25]. The cell wall peptidoglycan was obtained and purified according to the description by Schleifer & Kandler [26]. Menaquinones were extracted and purified from freeze-dried cells and analysed as described previously using LC-MS [27]. Polar lipids were extracted and separated using a two-dimensional TLC and silica gel 60 F 254 aluminium-backed thin-layer plates (Merck) [28].

IDENTIFICATION OF ORGANIC PHOSPHATE SOLUBILIZING ABILITY OF STRAIN

The isolated and purified bacteria were prepared into bacterial suspensions and cultured overnight. Suspensions (10 μL) were absorbed and inoculated in the organic phosphus agar medium [29] with 3 replicates for each bacterium, and cultured at 30 ℃. On day 7, the medium was observed for the

appearance of transparent circles, which indicated that it had the function of dissolving organic phosphorus (Fig. S5).

RESULTS AND DISCUSSION

PHYLOGENETIC ANALYSIS

The complete nucleotide sequence (1426 bp, accession number: OP795868) was determined by cloning and sequencing, and the results of EzTaxon server analysis indicated that strain MI7-26^T exhibited the highest 16S rRNA gene sequence similarities with *Arthrobacter bambusae* GM18^T (98.57% sequence similarity), *Arthrobacter gyeryongensis* DCY72^T (98.48%), *Pseudarthrobacter enclensis* NIO-1008^T (97.48%) and less than 97.50% for other type strains. The NJ, ML and MP trees showed that strain MI7-26^T was alone in one branch and don't cluster with other type strains (Fig. 1, S1 and S2).

The draft genome sequence of strain MI7-26^T was 4,813,939 bp including 78 contigs with N50 as 141,811 coding sequences and has been deposited at NCBI GenBank under accession number JAPFFT000000000. The DNA G + C content of strain MI7-26^T was calculated to be 63.1 mol% according to the genome sequence, which included in the scope of G + C content reported for the genus *Arthrobacter* [10]. To further clarify the phylogenetic position of strain MI7-26^T, genome-based analyses were conducted against closely related species, which involved calculations of digital DNA-DNA hybridization (dDDH), average nucleotide identity (ANI), and average amino acid identity (AAI) (Table S1). The results showed that the ANI values with the closest relatives, *Arthrobacter ramosus* (88.17%), *Arthrobacter gyeryongensis* (87.57%), *Arthrobacter bambusae* (87.19%), and *Arthrobacter methylotrophus* (86.22%), were all below the suggested species threshold of 95–96%. Similarly, the dDDH values (35.7%, 34.2%, 32.9%, and 31.2%, respectively) were well below the 70% species delineation threshold. The corresponding AAI values were 91.2%, 90.85%, 90.67%, and 89.8%. These collective genomic findings robustly support the proposal that strain MI7-26^T represents a novel species within the genus *Arthrobacter*.

For a more precise phylogenetic resolution, genome-based phylogenetic trees were reconstructed using both the TYGS (Fig. 2) platform and the AutoMLST pipeline (Fig. S3). In both trees, strain MI7-26^T consistently formed a distinct clade alongside *A. ramosus*, *A. gyeryongensis*, and *A. bambusae*, confirming its phylogenetic position within the genus *Arthrobacter* and its uniqueness as an independent lineage.

PHENOTYPIC AND PHYSIOLOGICAL CHARACTERISTICS

Cells of strain MI7-26^T were rod shaped of 0.4–0.7 μm in width and 1.0-1.6 μm in length (Fig. 3). The positive results of Voges-Proskauer, methyl red test, hydrolysis of Tween 80, enzyme activities of esterase (C4), esterase lipase (C8), lipase (C14), leucine arylamidase, valine arylamidase, cystine arylamidase, trypsin, chymotrypsin, acid phosphatase, naphthol-AS-BI-phosphohydrolase, *α*-

galactosidase, *β*-glucuronidase, *α*-glucosidase and *N*-acetyl-*β*-glucosaminidase, assimilation of glucose and mannose for strain MI7-26^T were different from *A. bambusae* GM18^T. The negative results of oxidase, hydrolysis of esculin, Tween 20, enzyme activity of *β*-galactosidase, assimilation of urease and *β*-glucosidase for strain MI7-26^T were different from *A. bambusae* GM18^T and *A. gyeryongensis* DCY72^T. Differential characteristics of strain MI7-26^T and the reference strains were shown in Table 1.

Table 1

Differential characteristics between strain MI7-26^T and the type strains of its closely related species Strains: 1, MI7-26^T; 2, *Arthrobacter bambusae* GM18^T; 3, *Arthrobacter gyeryongensis* DCY72 ^{T}; 4, *Pseudarthrobacter enclensis* NIO-1008^T. Data of strains 1 and 2 are from this study, and other data were obtained from Hoang et al. [5] and Dastager et al. [30], respectively. All strains were cultivated with the same medium and growth conditions. +, Positive; -, negative; w, weak reaction; nd, not determined.

Characteristic	**1**	**2**	**3**	**4**
Oxidase	-	+	+	-
Voges-Proskauer	+	-	nd	-
Methyl red test	+	-	nd	-
Hydrolysis of:				
Esculin	-	+	+	-
Cellulose	-	-	+	-
Gelatin	-	-	+	-
Tween 20	-	+	+	nd
Tween 80	+	-	-	nd
Enzyme activities:				
Alkaline phosphatase	-	-	+	-
Esterase (C4)	+	-	w	+
Esterase Lipase (C8)	+	-	+	-
Lipase (C14)	+	-	+	-
Valine arylamidase	+	-	+	-
Cystine arylamidase	+	-	+	-
Trypsin	+	-	+	-
Chymotrypsin	+	-	+	-
Naphthol-AS-BI-phosphohydrolase	+	-	+	+
α-Galactosidase	+	-	+	-
β-Galactosidase	-	+	+	-
β-Glucuronidase	+	-	-	-
α-Glucosidase	+	-	+	-

Characteristic	1	2	3	4
β-Glucosidase	+	+	+	-
N-Acetyl-*β*-glucosaminidase	+	-	-	-
Assimilation of:				
Urease	-	+	+	-
β-Glucosidase	-	+	+	-
β-Galactosidase	-	+	-	+
Glucose	+	-	+	+
Arabinose	-	-	-	+
Mannose	+	-	+	+
Mannitol	+	+	+	-
N-Acetyl-glucosamine	+	+	-	-
Maltose	-	-	+	+
Gluconate	-	-	+	+
DNA G + C content (mol%)	63.1	61.0	64.5	61.3

CHEMOTAXONOMIC ANALYSIS

The cellular fatty acids (> 1%) of strain MI7-26^T were anteiso-$C_{15:0}$ (71.9%), anteiso-$C_{17:0}$ (16.5%), iso-$C_{16:0}$ (4.5%), iso-$C_{15:0}$ (3.7%) and $C_{16:0}$ (1.1%), and other fatty acids shown in Table 2. Strain MI7-26^T shared a similar fatty acid profile with its close relatives *A. bambusae* GM18^T and *A. gyeryongensis* DCY72^T. In contrast, it was distinct from *P. enclensis* NIO-1008^T, which contained fatty acids such as iso-C15:0 and iso-C16:0 that were absent in MI7-26^T [30]. The cell-wall peptidoglycan consisted of alanine and lysine, and its structure was the A3*α* type. The predominant respiratory quinone of strain MI7-26^T was MK-9(H_2), which was similar to genus *Arthrobacter*. The major polar lipids had diphosphatidylglycerol, phosphatidylglycerol, phosphatidylinositol, two unidentified phospholipids, one unidentified aminolipid and two unidentified glycolipids (Fig. S4).

Table 2

Cellular fatty acids of strain MI7-26^T and its relative reference strains Strains: 1, MI7-26^T; 2, *Arthrobacter bambusae* GM18^T; 3, *Arthrobacter gyeryongensis* DCY72^T; 4, *Pseudarthrobacter enclensis* NIO-1008^T. Values are percentages of the total fatty acids. TR, Trace amount (< 0.5%).

Fatty acids	1	2	3	4
$C_{14:0}$	tr	tr	tr	1.9
$C_{16:0}$	1.1	3.4	1.0	5.5
$C_{18:0}$	tr	1.0	-	-
iso-$C_{14:0}$	0.5	1.7	1.5	2.0
iso-$C_{15:0}$	3.7	4.9	7.8	15.5
iso-$C_{16:0}$	4.5	7.9	6.6	10.5
iso-$C_{17:0}$	0.5	tr	0.7	2.1
anteiso-$C_{15:0}$	71.9	64.3	71.4	47.9
anteiso-$C_{17:0}$	16.5	14.2	10.2	10.2

Summed Features are fatty acids that cannot be resolved reliably from another fatty acid using the chromatographic conditions chosen. The MIDI system groups these fatty acids together as one feature with a single percentage of the total.

GENOME ANALYSES

The genome of strain MI7-26^T was 4,817,452 bp long including 114 contigs with an N50 value of 152,509 and a genome coverage of 80.0×. The DNA G + C content was 63.1 mol%, the mean length was 422,58.4bp. The MI7-26^T genome's RAST annotation provides initial annotations of gene functions for MI7-26^T's genomes, such as phosphorus metabolism (consistent with its observed ability to solubilize organic phosphorus, Fig. S5), secondary metabolism, etc. Furthermore, we annotated the genome of MI7-26^T through Proksee (https://proksee.ca/projects/new accessed on 28 Oct 2025) (Fig. S6). Antibiotic resistance gene prediction identified the *vanY* gene, which is part of the *vanB* cluster. This gene cluster confers resistance to glycopeptide antibiotics (https://card.mcmaster.ca/ accessed on 28 Oct 2025).

TAXONOMIC CONCLUSION

On the basis of the phenotypic, phylogenetic and chemotaxonomic characteristics described above, strain MI7-26^T is considered to represent a novel species within the genus *Arthrobacter* for which the name *Arthrobacter rhizophilus* sp. nov. is proposed.

DESCRIPTION OF ARTHROBACTER RHIZOPHILUS SP.NOV

Arthrobacter rhizophilus (rhi.zo'phi.lus. Gr. fem. n. *rhiza*, a root; Gr. masc. adj. *philos*, loving; N.L. masc. adj. *rhizophilus*, root-loving)

Cells are aerobic, Gram-staining-positive, rod-shaped, non-motile and 0.4–0.6 μm in width and 1.0-1.6 μm in length. The colonies are 1.0-2.4 mm in diameter, white, transparent and have regular margins and smooth after growth at 30 ℃ for 3 days on NA medium. The temperature range for growth is from 25 to 37 ℃ (optimum, 30 ℃). The pH range for growth is from 6.0 to 10.0 (optimum, 7.0). The NaCl concentration range for growth is from 0 to 6.0% (optimum, 0.5%).

Produce catalase and acetoin, and hydrolyzes Tween 80, but does not produce oxidase or hydrogen sulfide, and does not hydrolyze starch, casein, gelatin, cellulose, Tween 20 or Tween 60. Produce esterase (C4), esterase lipase (C8), lipase (C14), leucine arylamidase, valine arylamidase, cystine arylamidase, trypsin, chymotrypsin, acid phosphatase, naphthol-AS-BI-phosphohydrolase, α-galactosidase, β-glucuronidase, α-glucosidase, β-glucosidase, N-acetyl-β-glucosaminidase, α-mannosidase and α-fucosidase, but not alkaline phosphatase or β-galactosidase. It assimilates glucose, mannose, mannitol and N-acetyl-glucosamine, and produces acid from D-ribose, D-galactose, D-glucose, D-fructose, D-mannose, D-maltose, D-lactose, D-sucrose and D-trehalose.

The major fatty acids (> 10%) are anteiso-$C_{15:0}$ and anteiso-$C_{17:0}$. The peptidoglycan structural type is A3α type. The major quinone is menaquinone MK-9(H_2). The main polar lipids are diphosphatidylglycerol, phosphatidylglycerol, phosphatidylinositol, two unidentified phospholipids, one unidentified aminolipid and two unidentified glycolipids.

The type strain is MI7-26^T (= GDMCC 1.3254^T = JCM 35693^T), which was isolated from felty soil collected from rhizosphere soil, Nyingchi, Xizang, China. The genomic DNA G + C content of the type strain is 63.1 mol%.

Abbreviations

ANI
average nucleotide identity
dDDH
digital DNA-DNA hybridization
AAI
average amino acid identity
NA
nutrient agar
NB
nutrient broth

Declarations

Author contributions

Conceptualization, funding acquisition and supervision: ZX and ZR; Laboratory work, data analysis and writing of the original draft: YW, CZ and DK; Article content revision, data organization, and formatting: YW; Reviewing and editing of the manuscript: ZX and ZR. All authors have read and approved the final version of the manuscript.

Funding information

This research was supported by the National Key Research and Development Program of China (2023YFE0104900) and the National Natural Science Foundation of China (NSFC 3211101206).

Data availability

The GenBank/EMBL/DDBJ accession number for the 16S rRNA gene sequence of strain MI7-26^T is OP795868. The GenBank accession number for the draft genome sequence of strain MI7-26^T is JAPFFT000000000 (RefSeq assembly accession: GCF_026241105).

Conflict of interest

The research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest. All the authors declare that they have no conflict of interest.

Ethical Approval

The authors have declared that no ethical issues exist.

Research Involving Human and/or Animal Participants

This article does not contain any studies with human participants or animals performed by any of the authors.

Consent to Participate and Consent for Publication

All authors agree to have participated in the research proposed to be published and agree to be published in the journal.

References

1. Lin JZ (1999) Rare alpine flowers—*Meconopsis*. Chin Flower Bonsai 7:49 (In Chinese)
2. Wu ZY (1990) Poppies, Citrus family. In Flora of China; Editorial Committee of the Flora of China, Chinese Academy of Sciences: Beijing, China, Volume 32. (In Chinese)
3. Wu X, Rensing C, Han D, Xiao KQ, Dai Y, Tang Z, Liesack W, Peng J, Cui Z, Zhang F (2022) between Soil Microbiomes mSystems 7:e0110721. https://doi.org/10.1128/msystems.01107-21. Genome-

Resolved Metagenomics Reveals Distinct Phosphorus Acquisition Strategies

4. Conn HJ, Dimmick I (1947) Soil bacteria similar in morphology to *Mycobacterium* and *Corynebacterium*. J Bacteriol 54:291–303
5. Hoang VA, Kim YJ, Nguyen NL, Yang DC (2014) *Arthrobacter gyeryongensis* sp. nov., isolated from soil of a *Gynostemma pentaphyllum* field. Int J Syst Evol Microbiol 64:420–425. https://doi.org/10.1099/ijs.0.053967-0
6. Fang Z, Zhao X, Wu Q, Li S, Liu Q, Tan L, Weng Q (2022) *Arthrobacter cavernae* sp. nov., a novel actinobacterium isolated from sediment of karst cave. Int J Syst Evol Microbiol 72:5445. https://doi.org/10.1099/ijsem.0.005445
7. Park Y, Kook M, Ngo HT, Kim KY, Park SY, Mavlonov GT, Yi TH (2014) *Arthrobacter bambusae* sp. nov., isolated from soil of a bamboo grove. Int J Syst Evol Microbiol 64:3069–3074. https://doi.org/10.1099/ijs.0.064550-0
8. Busse HJ (2016) Review of the taxonomy of the genus *Arthrobacter*, emendation of the genus *Arthrobacter sensu lato*, proposal to reclassify selected species of the genus *Arthrobacter* in the novel genera *Glutamicibacter* gen. nov., *Paeniglutamicibacter* gen. nov., *Pseudoglutamicibacter* gen. nov., *Paenarthrobacter* gen. nov. and *Pseudarthrobacter* gen. nov., and emended description of *Arthrobacter roseus*. Int J Syst Evol Microbiol 66:9–37. https://doi.org/10.1099/ijsem.0.000702
9. Yoon SH, Ha SM, Kwon S, Lim J, Kim Y et al (2017) Introducing EzBioCloud: A taxonomically united database of 16S rRNA and whole genome assemblies. Int J Syst Evol Microbiol 67:1613–1617. https://doi.org/10.1099/ijsem.0.001755
10. Thompson JD, Gibson TJ, Plewniak F, Jeanmougin F, Higgins DG (1997) The clustal x windows interface: flexible strategies for multiple sequence alignment aided by quality analysis tools. Nucleic Acids Res 25:4876–4882. https://doi.org/10.1093/nar/25.24.4876
11. Saitou N, Nei M (1987) The neighbor-joining method: a new method for reconstructing phylogenetic trees. Mol Biol Evol 4:406–425. https://doi.org/10.1093/oxfordjournals.molbev.a040454
12. Felsenstein J (1981) Evolutionary trees from DNA sequences: a maximum likelihood approach. J Mol Evol 17:368–376. https://doi.org/10.1007/BF01734359
13. Kumar S, Stecher G, Tamura K (2016) mega 7: Molecular evolutionary genetics analysis version 7.0 for bigger datasets. Mol Biol Evol 33:1870–1874. https://doi.org/10.1093/molbev/msw054
14. Meier-Kolthoff JP, Göker M (2019) TYGS is an automated high-throughput platform for state-of-the-art genome-based taxonomy. Nat Commun. https://doi.org/10.1038/s41467-019-10210-3
15. Alanjary M, Steinke K, Ziemert N (2019) AutoMLST: an automated web server for generating multi-locus species trees highlighting natural product potential. Nucleic Acids Res 47:W276–W282. https://doi.org/10.1093/nar/gkz282
16. Bankevich A, Nurk S, Antipov D, Gurevich AA, Dvorkin M et al (2012) SPAdes: a new genome assembly algorithm and its applications to single-cell sequencing. J Comput Biol 19(5):455–477. https://doi.org/10.1089/cmb.2012.0021

17. Riesco R, Trujillo ME (2024) Update on the proposed minimal standards for the use of genome data for the taxonomy of prokaryotes. Int J Syst Evol Microbiol 74:006300. https://doi.org/10.1099/ijsem.0.006300
18. Meier-Kolthoff JP, Auch AF, Klenk H-P, Göker M (2013) Genome sequence-based species delimitation with confidence intervals and improved distance functions. BMC Bioinformatics 14:60. https://doi.org/10.1186/1471-2105-14-60
19. Yoon SH, Ha SM, Lim JM, Kwon SJ, Chun J (2017) A large-scale evaluation of algorithms to calculate average nucleotide identity. Antonie Van Leeuwenhoek 110:1281–1286. https://doi.org/10.1007/s10482-017-0844-4
20. Han C, Shi CP, Liu LM, Han JC, Yang QQ et al (2024) Majorbio Cloud 2024: Update single-cell and multiomics workflows. https://doi.org/10.1002/imt2.217. Imeta e217
21. Ruan ZY, Wang YW, Song JL, Jiang SH, Wang HM et al (2014) *Kurthia huakuii* sp. nov., isolated from biogas slurry, and emended description of the genus *Kurthia*. Int J Syst Evol Microbiol 64:518–521. https://doi.org/10.1099/ijs.0.056044-0
22. Skerman VBD (1967) A Guide to the Identification of the Genera of Bacteria, 2nd edn. Williams & Wilkins, Baltimore. https://doi.org/10.1099/00207713-18-3-276
23. Smibert RM, Krieg NR (1994) Phenotypic characterization. In Methods for General and Molecular Bacteriology, pp. 607–654. Edited by P. Gerhardt, R. G. E. Murray, W. A. Wood & N. R. Krieg. Washington, DC: American Society for Microbiology
24. Sakamoto M, Suzuki M, Umeda M, Ishikawa I, Benno Y (2002) Reclassification of *Bacteroides forsythus* (Tanner 1986) as *Tannerella forsythensis* corrig., gen. nov., comb. nov. Int J Syst Evol Microbiol 52:841–849. https://doi.org/10.1099/00207713-52-3-841
25. MacKenzie SL (1987) Gas chromatographic analysis of amino acids as the N-heptafluorobutyryl isobutyl esters. J Assoc Off Anal Chem 70:151–160
26. Schleifer KH, Kandler O (1972) Peptidoglycan types of bacterial cell walls and their taxonomic implications. Bacteriol Rev 36:407–477. https://doi.org/10.1128/br.36.4.407-477.1972
27. Minnikin DE, O'Donnell AG, Goodfellow M, Alderson G, Athalye M et al (1984) An integrated procedure for the extraction of bacterial isoprenoid quinones and polar lipids. J Microbiol Methods 2:233–241. https://doi.org/10.1016/0167-7012(84)90018-6
28. Kates M (1986) Techniques of Lipidology, 2nd edn. Elsevier, Amsterdam. https://doi.org/10.1016/S0075-7535(08)70544-8
29. Xu XH, Zheng HY (1960) Chinese Academy of Sciences, Microbiology Department, Institute of Forestry and Soil Research. Handbook of Methods for Soil Microbiological Analysis. Science, Beijing, China
30. Dastager SG, Liu Q, Tang SK, Krishnamurthi S, Lee JC, Li WJ (2014) *Arthrobacter enclensis* sp. nov., isolated from sediment sample. Arch Microbiol 196:775–782. https://doi.org/10.1007/s00203-014-1016-9

Figures

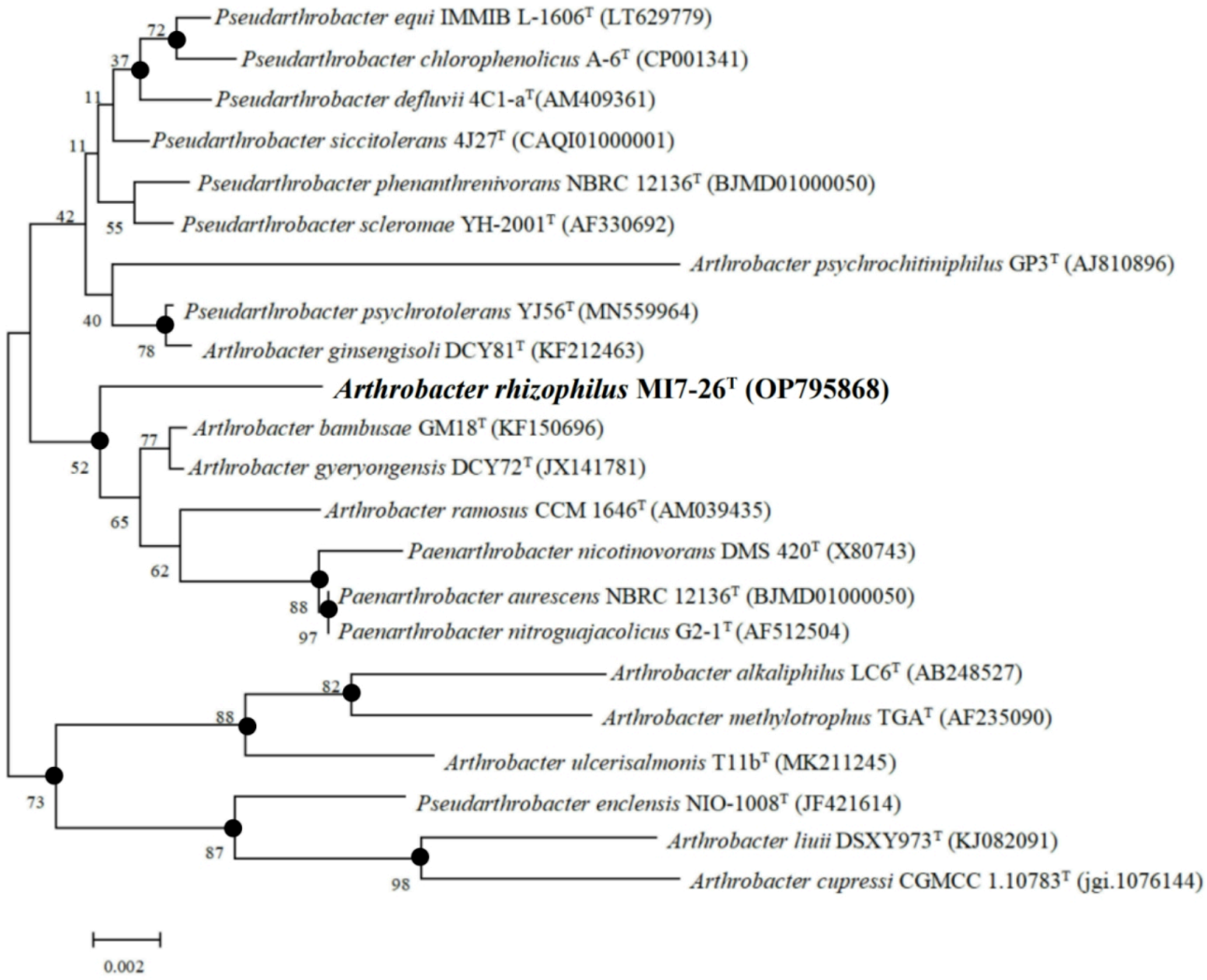

Figure 1

Phylogenetic tree based on neighbor-joining methods that displays the links between strain MI7-26^T and related taxa. Black circles represent nodes that were also present in the trees generated using the maximum-likelihood and maximum-parsimony methods. At the branch nodes, bootstrap values based on 1000 replicates are displayed. The scale bar represents 0.002 substitutions per nucleotide site.

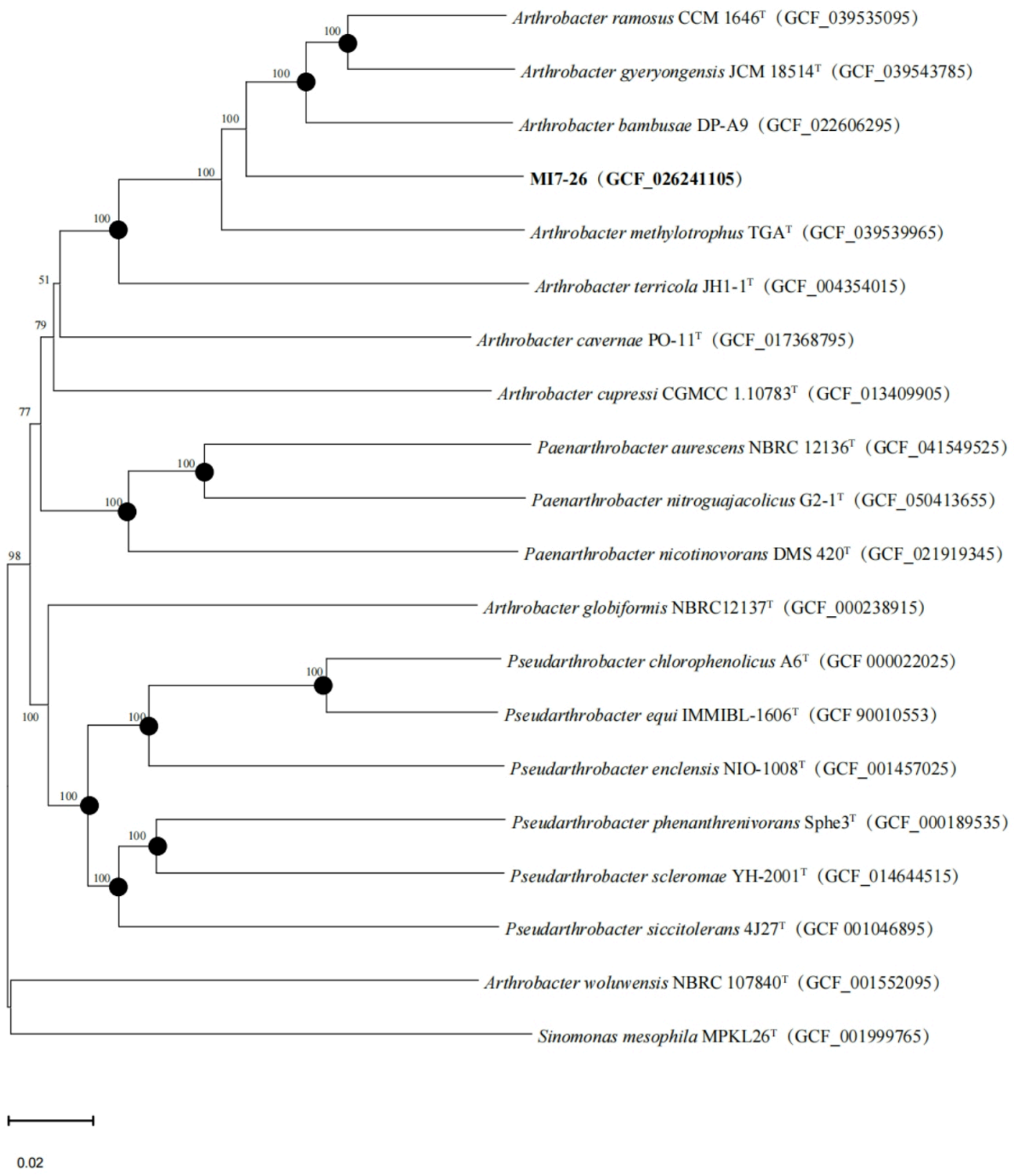

Figure 2

Phylogenetic tree was inferred from whole-genome comparative analysis, illustrating the relationship between strain MI7-26^T and its related species. The tree was constructed using the Type (Strain) Genome Server (TYGS; https://tygs.dsmz.de/). Black circles at the nodes indicate branches that were also recovered in the phylogeny generated using the AutoMLST pipeline

(https://automlst.ziemertlab.com). Bootstrap values based on 1000 replicates are shown at the branch nodes. The scale bar represents 0.02 substitutions per nucleotide site.

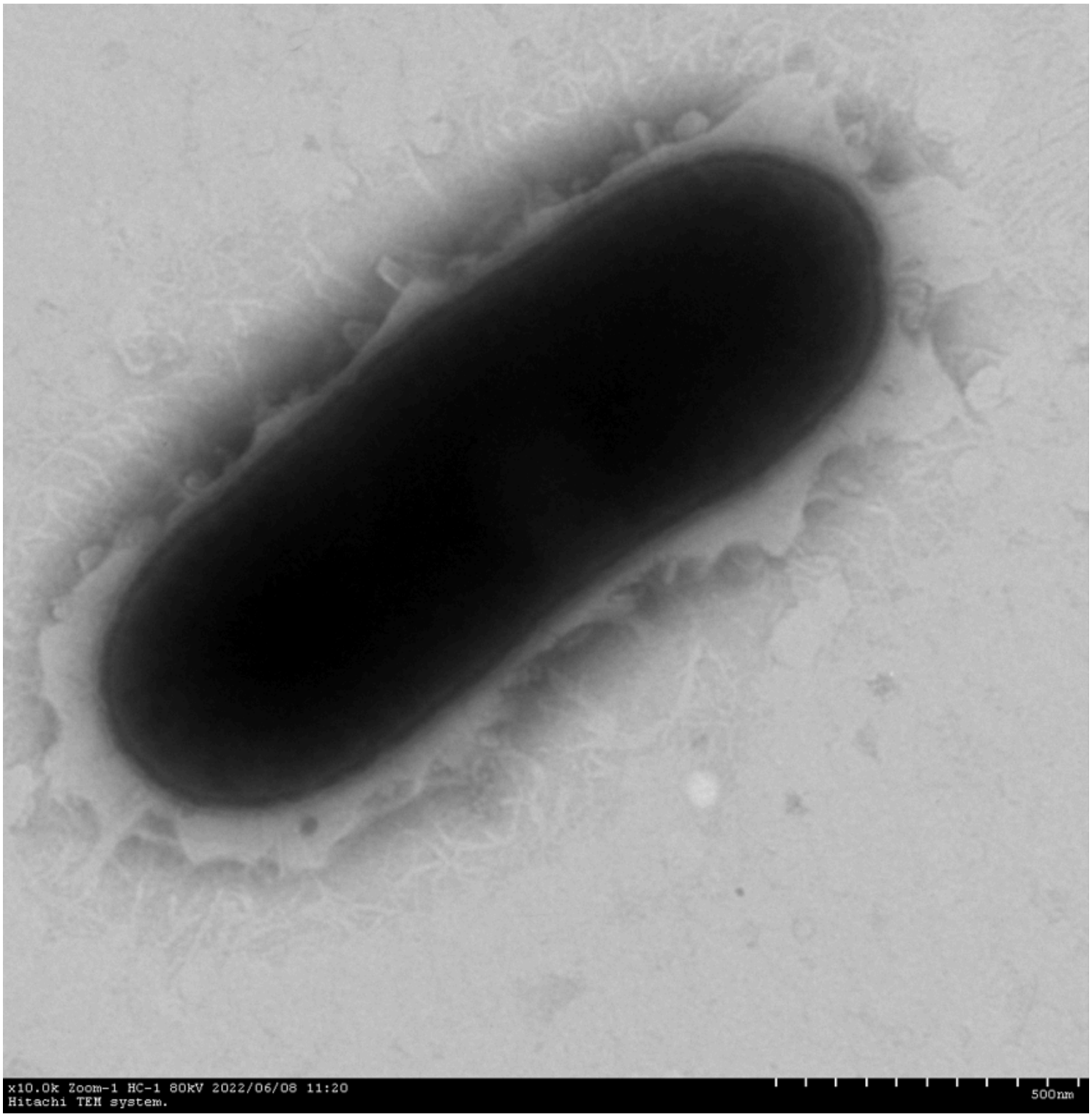

Figure 3

Transmission electron micrograph of cell of strain MI7-26^{T}. Bar, 1.0 μm. The strain was incubated on NA at 37 ℃ for 3 days.

Supplementary Files

This is a list of supplementary files associated with this preprint. Click to download.

- Supplementarymaterials.docx