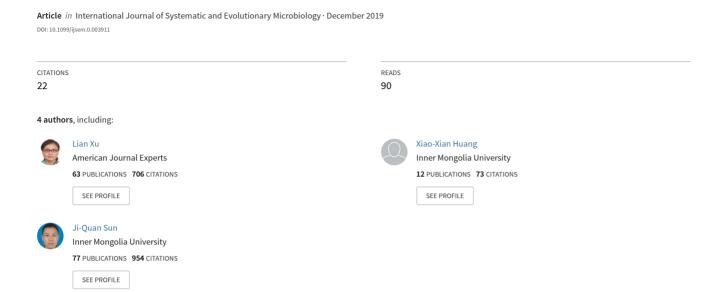
Lysobacter alkalisoli sp. nov., a chitin-degrading strain isolated from saline-alkaline soil



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TAXONOMIC DESCRIPTION

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Lysobacter alkalisoli sp. nov., a chitin-degrading strain isolated from saline-alkaline soil

Lian Xu, Xiao-Xian Huang, De-Liang Fan and Ji-Quan Sun*

Abstract

Strains of *Lysobacter*, thought to play vital roles in the environment for their high enzyme production capacity, are ubiquitous in various ecosystems. During an analysis of bacterial diversity in saline soil, a Gram-stain-negative, aerobic, chitin-degrading bacterial strain, designated SJ-36^T, was isolated from saline-alkaline soil sampled at Tumd Right Banner, Inner Mongolia, PR China. Strain SJ-36^T grew at 4–40 °C (optimum, 30 °C), pH 5.0–10.0 (optimum, pH 7.0–8.0) and 0–6% NaCl (optimum, 1.0%). Oxidase and catalase activities were positive. A phylogenetic tree based on 16S rRNA gene sequences and the phylogenomic tree both showed that strain SJ-36^T formed a tight clade with *Lysobacter maris* KMU-14^T (sharing 97.6% 16S rRNA gene similarity) and *Lysobacter aestuarii* S2-C^T (97.8%). The major polar lipids of strain SJ-36^T were phosphatidylethanolamine, diphosphatidylgycerol, phosphatidylgycerol, two unidentified lipids and one unidentified phospholipid. The major fatty acids were iso-C_{15:0} (37.5%), summed feature 9 (14.0%; iso-C_{17:1} ω 9c and/or C_{16:0} 10-methyl) and iso-C_{11:0} (10.6%). Q-8 was the predominant ubiquinone. Its genomic DNA G+C content was 66.6 mol%. The average nucleotide identity values of strain SJ-36^T to *L. maris* KMU-14^T, *L. aestuarii* S2-C^T and other type strains were 81.5, 79.1 and <79.0%, respectively. The results of physiological, phenotypic and phylogenetic characterizations allowed the discrimination of strain SJ-36^T from its phylogenetic relatives. *Lysobacter alkalisoli* sp. nov. is therefore proposed with strain SJ-36^T (=CGMCC 1.16756^T=KCTC 43039^T) as the type strain.

The genus Lysobacter, belonging to the family Xanthomonadaceae of the Gammaproteobacteria, was first described by Christensen and Cook [1] and later amended by Park et al. [2]. Currently, there are 54 valid Lysobacter species (www. bacterio.net/lysobacter.html) [3], including the recently published species Lysobacter psychrotolerans [4], Lysobacter silvisoli [5], Lysobacter tabacisoli [6], Lysobacter caseinilyticus [7], Lysobacter helvus, Lysobacter xanthus [8] and Lysobacter oculi [9]. They were isolated from various habitats, i.e., activated sludge, freshwater sediment, rhizosphere, soil and spongin [10]. Members of the genus Lysobacter are Gramnegative, aerobic rods that have a high genomic DNA G+C content (61.7-70.7 mol%) and contain ubiquinone 8 (Q-8) as the major respiratory quinone [10]. All validly named species of the genus Lysobacter show negative activities for urease and indole production [7]. Many strains of Lysobacter can produce enzymes and secondary metabolites [11]. Because strains of the genus live in widespread environments and have diverse physiological functions in the ecosystem [12], the isolation and identification of *Lysobacter* strains from saline soil may help us to understand its role in this environment. Here, we reported the polyphasic characterization of a Gram-negative, aerobic bacterium, designated SJ-36^T, which was isolated from saline-alkaline soil during an investigation into the saline-alkaline soil microbial community.

The saline-alkaline soil used to isolate strains was collected from farmland in Tumd Right Banner (110° 49′ 15″ E 40° 35′ 55″ N), Inner Mongolia, PR China. Briefly, the bacterial strains were isolated using the 10-fold dilution plating technique on Luria–Bertani (LB) agar (10.0 g l $^{-1}$ tryptone, 5.0 g l $^{-1}$ yeast extract, 10.0 g l $^{-1}$ NaCl and 20 g l $^{-1}$ agar; pH 7.0). After 5 days of incubation at 30 °C in the dark, the cultured colonies were selected and purified by repeated streaking on LB agar. The ability of these strains to degrade chitin or cellulose was

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Keywords: Lysobacter alkalisoli sp. nov.; saline soil; polyphasic analysis; chitin; degradation.

Abbreviations: ANIb, the average nucleotide identity based on BLASTing; KCTC, the Korean Collection for Type Cultures; LB, Luria-Bertani. The GenBank/EMBL/DDBJ accession numbers for the 16S rRNA gene and the complete genome sequence of strain SJ- 36^{T} are MK641452 and CP041242, respectively. The Whole Genome Shotgun projects for strains L. Maris KMU- 14^{T} and L. M0 accessions VICD000000000 and VICE000000000, respectively.

Five supplementary figures and five supplementary tables are available with the online version of this article.

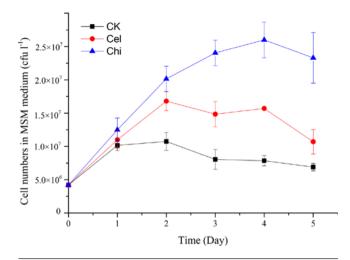


Fig. 1. Growth of strain SJ-36^T in minimal salt medium (0.5 mg l^{-1} yeast extract) supplemented with 100 mg l^{-1} cellulose (circle) or chitin (triangle). The inoculum size is 0.1% (m/m; approximately $4.2 \times 10^6 \, \text{c.f.u.}^{-1}$), CK (control; square) was only inoculated strain SJ-36^T.

checked according to a previously described method [13]. One of the designated strains, SJ-36^T, which was able to degrade chitin, was isolated. The analysis revealed that strain SJ-36^T could grow in minimal salt medium (0.5 g l⁻¹ KH₂PO₄, 1.5 g l⁻¹ K₂HPO₄, 1.0 g l⁻¹ NH₄NO₃, 1.0 g l⁻¹ NaCl, 1.0 mg l⁻¹ yeast extract) with chitin supplemented as the major carbon and energy source (Fig. 1), suggesting that strain SJ-36^T could degrade chitin. Therefore, strain SJ-36^T may play a vital role in the carbon cycle in saline soils.

After growth in LB broth at 30 °C for 2 days in the dark, cells of strain SJ-36^T were harvested for extraction of the genomic DNA and amplification of the 16S rRNA gene by following a previously described protocol [14]. After ligation into the pMD19-T vector (TaKaRa) following the manufacturer's instructions, the amplified 16S rRNA gene fragment was sequenced. One almost-complete 16S rRNA gene sequence (1421 nt) was compared with the DNA sequences available in GenBank using BLAST (http://blast.ncbi.nlm.nih.gov) to determine the approximate taxonomic affiliation of the strain. The similarities between strain SJ-36^T and its relatives were evaluated by using the EzTaxon-e server (www.ezbiocloud.net) [15]. After multiple alignment of the data using CLUSTAL_X software [16], phylogenetic analysis was performed using the software package меда version 6.1 [17]. Phylogenetic trees were then reconstructed with the neighbour-joining [18], maximum-likelihood [19] and minimum-evolutionary [20, 21] algorithms. The tree topology was assessed by the bootstrap resampling method with 1000 replicates [22]. The phylogenetic analysis showed that strain SJ-36^T formed a stable clade with *Lysobacter maris* KMU-14^T (=KCTC 42381^T; 97.6%) [23] and Lysobacter aestuarii S2-C^T (=JCM 31130^T; 97.8%) (Fig. 2, S1 and S2 available in the online version of this article) [24]. However, all the 16S rRNA gene similarities of strain SJ-36^T to valid type strains were below 98.7%, the threshold value for proposing a novel species [25]. Therefore,

L. maris KMU-14^T and *L. aestuarii* S2-C^T were selected as the reference strains for further tests and were obtained from the Korean Collection for Type Cultures (KCTC) and the Japan Collection of Microorganisms (JCM), respectively.

For whole-genome sequencing, the genomic DNA of strains SJ-36^T, L. maris KMU-14^T and L. aestuarii S2-C^T were prepared by a modified SDS-based DNA extraction method [14]. The complete genome of strain SJ-36^T was sequenced using a combination of Hiseq2500 (Illumina) and Pacific Biosciences Sequel sequencing platforms, and the pairedend sequence data from the Illumina platform was used to proofread the PacBio assembly sequence as described earlier [13]. The draft genomic sequences of L. maris strain KMU- 14^{T} and L. aestuarii strain S2-CT were sequenced by using the Illumina Hiseq2500. The authenticity and contamination of these two genome sequences were checked using 16S rRNA and/or gyrase subunit B (gyrB) genes using BLAST. The genome sequence of strain SJ-36^T was annotated using the RAST server [26] with default parameters. The average nucleotide identity based on BLAST (ANIb) of strain SJ-36^T to L. maris KMU-14^T, L. aestuarii S2-C^T and the other type strain of Lysobacter (Table S1) were calculated using JspeciesWS online (http://jspecies.ribohost.com/jspeciesws/#analyse) [27]. To reconstruct a phylogenomic tree, the whole genome sequences of *Lysobacter* strains were retrieved from GenBank. After predicting the amino sequences using Prodigal version 2.6.3 [28], a phylogenomic tree based on all predicted amino sequences was reconstructed using standalone software CVtree 3.0 with the default parameters [29, 30]. The genome of strain SJ-36^T was also compared to those of *Lysobacter* strains by using Mauve Genome Alignment version 2.3.1 with the progressiveMauve algorithm [31]. The complete genome of strain SJ-36^T consisted of a circular chromosome, which was 3857091 bp long and had a G+C content of 66.6 mol% (Fig. 3, Tables 1 and S1). A total of 49 tRNA genes and two rRNA operons were predicted by the Prodigal software. The G+C content of strain SJ-36^T was located in the range of 61.7–70.7 mol% described for the genus Lysobacter [10]; however, the value was much lower than those of L. maris KMU-14^T (68.6 mol%) and *L. aestuarii* S2-C^T (69.4 mol%), as well as the other *Lysobacter* type strains (Tables 1 and S2). RAST predicted that the genome of strain SJ-36^T contained 3379 coding sequences with an average size of 1011 bp, giving a gene density of 84% (Fig. S3). The ANIb of strain SJ-36^T to L. maris KMU-14^T, L. aestuarii S2-C^T and other closely related Lysobacter type strains were 81.5, 79.1 and <79.0%, respectively. This is much lower than the threshold value of 95–96% for proposing a novel species [32]. It is noteble that the average ANIb of strain SJ-36^T to its closely related *Lysobacter* strains was slightly higher than those of the nearest relatives (Table S3). The phylogenomic tree revealed that strain SJ-36^T was also clustered with L. maris KMU-14^T and L. aestuarii S2-C^T (Fig. 4). The genome of strain SJ-36^T was compared to those of the nearest *Lysobacter* species. The Mauve results showed that strain SJ-36^T has high synteny with these seven genome strains, even though it exhibited a greater number of rearrangements, insertions and/or deletions (Fig. S4). In

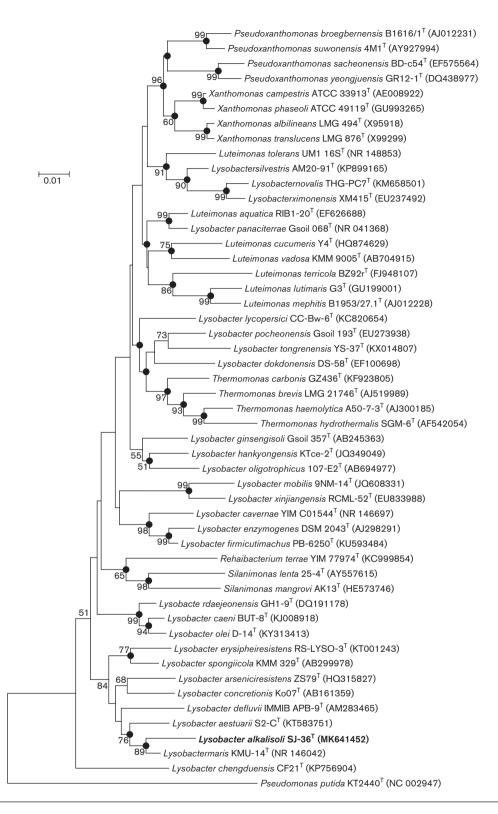


Fig. 2. Phylogenetic tree reconstructed by the neighbour-joining method based on the 16S rRNA gene sequences. Bootstrap values (numbers on branch nodes are expressed as percentages of 1000 replications) >50% are shown at branch points. Filled circles indicate that the corresponding nodes were recovered in the minimum-evolution and maximum-likelihood trees. Bar, 0.01 substitutions per nucleotide position.

- Energy production & conversion
- Carbohydrate transport &metabolism
- Amino acid transport &metabolism
- Nucleotide transport &metabolism
- Coenzyme transport &metabolism
- Lipid transport &metabolism
- Inorganic ion transport &metabolism
- Secondary metabolites biosynthesis, transport & catabolism
- Translation, ribosomal structure & modification
- RNA procession & modification
- Transcription
- Replication, recombination & repair
- Chromatin structure &dynamics

- Cell cycle control, cell division, chromosome partitioning
- Nuclear structure
- Defense mechanisms
- Signal transduction mechanism
- Cell wall/membrane/envelope biogenesis
- Cell motility
- Cell mounty
- Cytoskeleton
- Extracellular structures
- Intracellular trafficking, secretion & vesicular transport
- Posttranslational modification, protein turnover, chaperones
- General function prediction only
- Function unknown
- No COG assignment

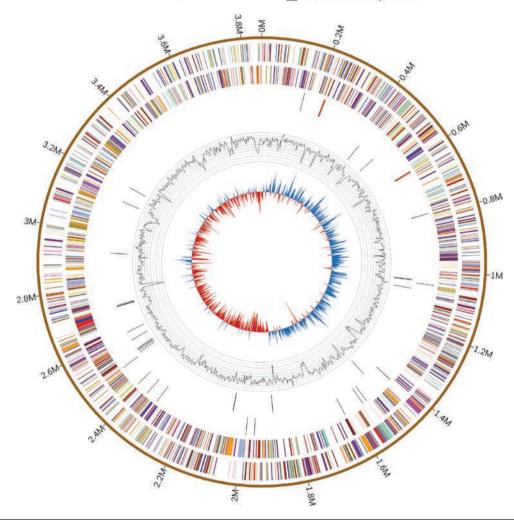


Fig. 3. Circular chromosome genome (A) of strain $SJ-36^T$. Position 1 of the chromosome was assigned to the first nucleotide of the *dnaA* gene. Ring 1 (from the outside) indicate the genome size, Ring 2 and Ring 3 indicate the CDS in forward and reverse strands, respectively. Ring 4 and Ring 5 show the tRNA (black) and rRNA (red) genes on the forward and reverse strands, respectively. Rings 6 and 7 indicate the G+C content, and GC skew [(C-G)/(C+G)], respectively. The colours of the genes indicate the COG categories, as indicated in the figure. The circular genome map was generated by Circos version 0.64 (http://circos.ca/).

Table 1. Differentiating characteristics of strain SJ-36^T compare to closely related strains

Strains: 1, SJ-36 $^{\text{T}}$; 2, Lysobacter maris KMU-14 $^{\text{T}}$; 3, Lysobacter aestuarii S2-C $^{\text{T}}$; 4, Lysobacter defluvii IMMIB APB-9 $^{\text{T}}$; 5, Lysobacter concretionis Ko07 $^{\text{T}}$; 6, Lysobacter arseniciresistens ZS79 $^{\text{T}}$; 7, Lysobacter spongiicola KMM 329 $^{\text{T}}$; 8, Lysobacter erysipheiresistens RS-LYS0-3 $^{\text{T}}$; 9, Lysobacter enzymogenes DSM 2043 $^{\text{T}}$. All strains are positive for catalase and alkaline phosphatase. All strains are negative for indolent production, α -galactosidase, β -glucuronidase, and β -galactosidase, and for assimilation of L-arabinose. Data for strains 4–9 were compiled from published literature [23, 24, 41–45]. +, positive; –, negative; w, weak; ND, no data.

Characteristics	1	2	3	4	5	6	7	8	9
pH range (optimum) for growth	5-10 (7-8)	6-8 (7)	5.5-9.0 (6.5-7.5)	ND	ND (6.8–7.5)	5-9 (7)	5.5-9.5 (6.5-8.5)	6-8	ND
Temperature range (optimum) for growth (°C)	4-40 (30)	20-40 (30)	15-40 (30)	22-37 (ND)	ND (25–30)	4-37 (28)	5-41 (25-28)	10-35	ND
Growth with NaCl range (%,w/v)	0-6.0	0-5.0	0-7.0	0-6.0	ND	0-4.0	0-6.0	0-5	0-2
Oxidase activity	+	-	+	+	+	+	+	+	+
Aesculin hydrolysis	+	+	+	-	-	_	-	ND	+
Esterase (C4)	+	+	+	ND	ND	+	+	-	ND
Esterase lipase (C8)	+	+	+	ND	ND	+	+	+	ND
Cystine arylamidase	-	-	-	-	ND	_	-	+	ND
β -Glucuronidase	-	-	-	-	-	_	-	-	+
Nitrate reduction	+	-	+	+	-	-	-	-	_
Arginine dihydrolase	+	+	-	-	-	_	-	-	_
Urease	+	+	-	+	-	_	-	-	_
Gelatin hydrolysis	_	+	+	+	+	+	+	+	+
Lipase (C14)	_	_	_	ND	ND	+	-	_	+
Leucine arylamidase	+	+	+	_	-	+	+	+	_
Valine arylamidase	+	_	+	ND	ND	+	-	w	ND
Trypsin	+	+	_	ND	ND	+	-	+	+
Chymotrypsin	+	-	+	ND	ND	+	+	W	ND
Acid phosphatase	+	+	+	ND	ND	+	+	W	ND
Naphthol-AS-BI-phosphohydrolase	+	+	+	ND	ND	+	+	+	ND
α-Glucosidase	_	+	_	_	_	_	_	_	+
N -Acetyl- β -glucosaminidase	+	_	+	_	_	_	_	_	_
α-Mannosidase	_	_	_	ND	ND	_	_	_	ND
α-Fucosidase	_	_	_	ND	ND	_	_	_	ND
Carbon assimilation:									
D-Glucose	+	+	+	_	_	_	_	+	+
D-Mannose	+	+	_	_	_	_	_	_	+
N-Acetyl-glucosamine	+	+	+	_	_	_	_	_	+
Maltose	+	+	+	_	_	_	_	_	+
Malic acid	_	_	_	_	_	_	_	_	+
Mannitol	_	_	_	_	_	_	_	_	+
Gluconate	_	_	_	_	W	_	_	_	_
Capric acid	_	_	_	_	_	_	_	_	ND

Continued

Table 1. Continued

Characteristics	1	2	3	4	5	6	7	8	9
Adipic acid	-	-	-	-	W	-	-	-	-
Phenylacetic acid	-	_	-	-	_	_	-	W	-
Trisodium citrate	-	-	-	+	-	-	-	-	+
DNA G+C content (mol%)*	66.6	68.6	69.4	70.3	67.2	69.6	67.6	68.0	69.9

^{*}Data for *L. erysipheiresistens* RS-LYSO-3^T was determined via the thermal denaturation method, while the others were calculated from the whole genome sequences.

addition, the genome analysis revealed that several genes involved into the transportation, degradation and regulation of chitin and its monomer were present in strain SJ-36^T (Table S4), suggesting that strain SJ-36^T was able to degrade chitin, even though the degradation rate of chitin was slow (Fig. 1). Among them, the chitinase gene has the highest similarity to *L. maris* strain HZ9B (CP029843). The genome sequence analysis supports that strain SJ-36^T is within the genus *Lysobacter*, but different from current valid species of *Lysobacter*.

For determining the cellular fatty acids, cells of strain SJ-36^T as well as its two relatives (L. maris KMU-14^T and L. aestuarii S2-C^T) were grown on trypticase soy agar (TSA; Difco) at 30°C and were harvested at roughly the same growth stage in the exponential growth phase (24h). The fatty acids were prepared and identified following the instructions of the Microbial Identification system (MIDI) as described previously [31]. Polar lipids were extracted and examined by two-dimensional TLC as described previously [33]. Menaguinones were extracted with chloroform/methanol (2:1, v/v) solution and analysed as described by Komagata and Suzuki [34] using HPLC. The cellular fatty acid profile of strain SJ-36^T was characterized by the fatty acids iso-C_{15:0} (37.5%), summed feature 9 (14.0%; iso- $C_{17:1}\omega 9c$ and/or $C_{16:0}$ 10-methyl) and iso- $C_{11:0}$ (10.6%). This fatty acid profile was in agreement with those of its two related Lysobacter type strains, although the relative abundance of some fatty acids were different (Table S5) [24]. The predominant isoprenoid quinone of strain SJ-36^T was Q-8, which is similar to L. maris KMU-14^T [23] and L. aestuarii S2-C^T [24]. The polar lipids of strain SJ-36^T consisted of phosphatidylethanolamine (PE) diphosphatidylglycerol (DPG), phosphatidylglycerol (PG), two unidentified lipids (Ls) and an unidentified phospholipid (PL). Strain SJ-36^T and its two closest relatives exhibited very similar lipid profiles, and they all had PE, DPG, PG and L1. However, PL2 was present in *L. maris* KMU-14^T and *L. aestu*arii S2-C^T, but absent in strain SJ-36^T; L2 was present in strain SJ-36^T, but absent in the other two strains; L3 was present in strain L. aestuarii S2-C^T, but absent in L. maris KMU-14^T and SJ-36^T; PL1 was present in SJ-36^T and L. aestuarii S2-C^T, but absent in L. maris KMU-14 $^{\rm T}$ (Fig. S5). The cellular fatty acid and polar lipid profiles both supported the affiliation of strain SJ-36^T to the genus *Lysobacter*.

After strain SJ-36^T was grown on LB agar at 30 °C for 2 days, cells were harvested, air-dried and negatively stained with 1%

(w/v) phosphotungstic acid. The morphological features of the strain were examined using transmission electron microscopy (JEM-1400, JEOL). Gram-staining and endospore formation were investigated as described by Smibert and Krieg [35]. The temperature range for growth was determined in LB broth at 4, 10, 15, 20, 25, 30, 35, 37, 40 and 45 °C. Growth at different pH values (pH 4.0-11.0 in 1.0 unit intervals) was assessed in minimal salt medium adjusted with 10% of HCl or NaOH solutions after 2 days of incubation at 30 °C. NaCl tolerance was tested using a modified LB broth amended with 0, 0.5, 1, 2, 3, 4, 5, 6, 7, 8 and 9% NaCl. Cell motility was determined in LB medium containing 0.4% agar [36]. The cell gliding motility was determined on LB agar by following a previous protocol [37]. Anaerobic growth was tested in LB medium supplemented with nitrate or sulfate as described by Kim et al. [38]. Flexirubin pigments were detected by covering the colonies with 20% KOH [39]. Antibiotic sensitivity tests were performed using the diffusion method [40] on LB agar with filter-paper discs (8 mm diameter) containing one of the following antibiotics: penicillin (10 U), oxazocilline (1 µg), ampicillin (10 μg), carbenicillin (100 μg), piperacillin (100 μg), cephalexin (30 µg), cefazolin (30 µg), cefradine (30 µg), cefuroxime (30 μg), ceftazidime (30 μg), cefatriaxone (30 μg), cefoperazone (75 µg), amikacin (30 µg), gentamycin (10 µg), kanamycin (30 μg), neomycin (30 μg), tetracycline (30 μg), doxycycline (30 µg), minocycline (30 µg), erythromycin (15 μg), norfloxacin (10 μg), medemycin (30 μg), ofloxacin (5 μg), ciprofloxacin (5 μg), vancomycin (30 μg), polymyxin B (30 μg), furazolidone (300 μg), chloramphenicol (30 μg) or clindamycin (30 µg). Oxidase and catalase activities were evaluated by adding oxidase reagent (bioMérieux) and 3% hydrogen peroxide solution to the fresh colony [36]. Other biochemical characteristics were tested using API 20NE, API ZYM and API 50CH kits (bioMérieux) according to the manufacturer's instructions. The reference strains L. maris KMU-14^T and L. aestuarii S2-C^T were simultaneously tested under the same conditions. Cells of strain SJ-36^T were Gram-stain-negative and rod-shaped. Cells of strain SJ-36^T were $0.5-1.0\,\mu m$ wide and $2.0-3.4\,\mu m$ long (Fig. 5). The other phenotypic features of strain SJ-36^T are listed in the species description and in Table 1. Strain SJ-36^T was positive for activities of catalase, oxidase and alkaline phosphatase; whereas it was negative for indole production, α -galactosidase, β -glucuronidase and β -galactosidase; these results are similar

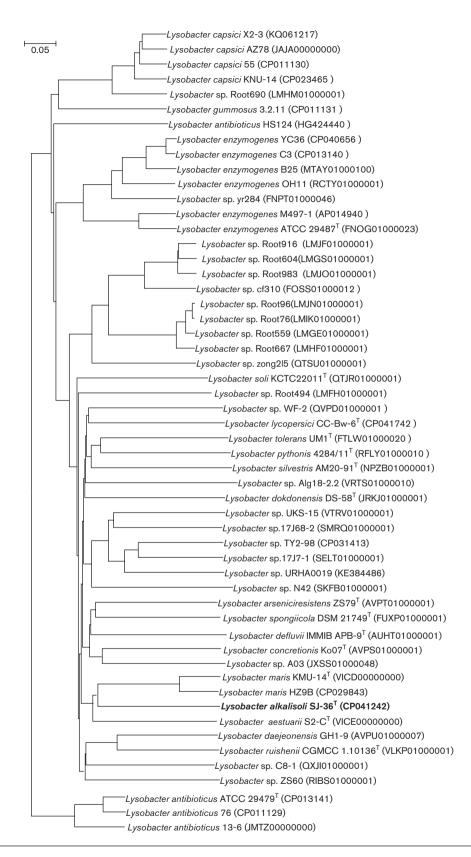


Fig. 4. Phylogenomic tree of *Lysobacter* strains reconstructed using the standalone software CVtree version 3.0 with the default parameters [29, 30].

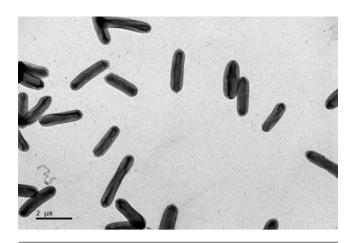


Fig. 5. Transmission electron microscope image of cells of strain SJ- 36^T , grown on LB agar for 24h at 30 °C. Bar, $2 \mu m$

to those for most other *Lysobacter* strains (Table 1). Besides the consistent characteristics with other Lysobacter strains, there are also different characteristics between SJ-36^T and the other Lysobacter strains. For example, compared to other Lysobacter strain, strain SJ-36^T could grow at a wider pH range and temperature range; strain SJ-36^T was found to be negative for gelatin hydrolysis and negative for N-acetyl-βglucosaminidase; the other eight *Lysobacter* strains (except *L*. *aestuarii* S2-C^T for *N*-acetyl-β-glucosaminidase), by contrast, were found to be negative for gelatin hydrolysis and positive for N-acetyl-β-glucosaminidase. Strain SJ-36^T was resistant to penicillin, oxazocilline, cefradine, medemycin and furazolidone; and sensitive to ampicillin, carbenicillin, piperacillin, cephalexin, cefazolin, cefuroxime, ceftazidime, cefatriaxone, cefoperazone, amikacin, gentamycin, kanamycin, neomycin, tetracycline, doxycycline, minocycline, erythromycin, norfloxacin, ofloxacin, ciprofloxacin, vancomycin, polymyxin B, chloramphenicol and clindamycin.

On the basis of the phylogenetic, phenotypic and genotypic data presented here, we propose that strain SJ-36^T should be classified as the type strain of a novel species within the genus *Lysobacter*, for which the name *Lysobacter alkalisoli* sp. nov. is proposed.

DESCRIPTION OF LYSOBACTER ALKALISOLI SP. NOV.

Lysobacter alkalisoli [al.ka.li.so'li. N.L. n. alkali (from Arabic al-qaliy), alkali; L. neut. n. solum soil; N.L. gen. n. alkalisoli of alkaline soil].

Cells are Gram-stain-negative, non-spore-forming, non-motile, rod-shaped, 0.5–1.0 µm wide and 2.0–3.4 µm long. Strain SJ-36^T can grow on LB agar and TSA. Colonies on LB agar at 30 °C for 48 h are circular with regular border, yellow-coloured, smooth, low convex and usually 2–3 mm in diameter. Grows at 4–40 °C (optimum, 30 °C), pH 5.0–10.0 (optimum, pH 7.0–8.0) and 0–6% NaCl (optimum, 1.0 %).

Positive for catalase, oxidase and hydrolysis of Tween 80, starch and chitin; negative for hydrolysis of cellulose and production of flexirubin-type pigments. In API 20NE tests, it is positive for activities to glucose fermentation, arginine dihydrolase, urease, nitrate reduction and aesculin hydrolysis, and negative for indole production and gelatin hydrolysis. In API ZYM tests, it is positive for alkaline phosphatase, esterase (C4), esterase lipase(C8), leucine arylamidase, valine arylamidase, trypsin, α-chymotrypsin, acid phosphatase, naphthol-AS-BI-phosphohydrolase and N-acetyl-β-glucosaminidase, and negative for lipase (C14), cystine arylamidase, α -galactosidase, β-galactosidase, β-glucuronidase, α-glucosidase, β-glucosidase, α-mannosidase and α-fucosidase. In API 50CH tests, it produces acid from mannitol, D-ribose, D-glucose, D-mannose, trehalose and D-tagatose and grows in mannitol, D-ribose, D-mannose, inositol, amygdalin, trehalose, D-lyxose, D-tagatose and D-arabinol. Strain SJ-36^T assimilates D-glucose, D-mannose, mannitol, maltose, D-ribose, pyruvate, sodium acetate, sodium succinate and N-acetyl-glucosamine; but does not assimilate malic acid, L-arabinose, gluconate, fructose, sucrose, capric acid, adipic acid, phenylacetic acid or trisodium citrate. Major polar lipids are diphosphatidylglycerol, phosphatidylglycerol, one unidentified phospholipid and two unidentified lipids. Q-8 is the major isoprenoid quinone. The main fatty acids are iso- $C_{15:0}$, summed feature 9 (iso- $C_{17:1}\omega 9c$ and/or C_{16:0} 10-methyl) and iso-C_{11:0}.

The type strain, SJ-36^T (=CGMCC 1.16756^T=KCTC 43039^T), was isolated from a top layer of saline-alkaline soil sampled in Inner Mongolia, PR China. The complete genome of the type strain consists of a circular chromosome, 3857091 bp long with a G+C content of 66.6 mol%.

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Conflicts of interest

The authors declare that there are no conflict of interests.

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