INTERNATIONAL JOURNAL OF SYSTEMATIC AND EVOLUTIONARY MICROBIOLOGY

TAXONOMIC DESCRIPTION

Xu *et al.*, *Int. J. Syst. Evol. Microbiol.* 2021;71:004593

DOI 10.1099/ijsem.0.004593

MICROBIOLOGY SOCIETY

Lysobacter penaei sp. nov., isolated from intestinal content of a Pacific white shrimp (*Penaeus vannamei*)

Shuaishuai Xu[1]†, Anzhang Li[1]†, Ming-Xia Zhang[1], Qing Yao[2] and Honghui Zhu[1,*]

Abstract

The polyphasic taxonomic approach was used to characterize a novel bacteria strain, designated SG-8^T, which was isolated from intestinal content of a Pacific white shrimp (*Penaeus vannamei*). Cells were Gram-stain-negative, aerobic, non-gliding rods. Growth occurred at 10–45 °C (optimum, 20–30 °C), pH 5.0–10.0 (optimum, 6.0–7.0) and in 0–6.0% (w/v) NaCl (optimum, 0–4.0%). The 16S rRNA gene sequence of strain SG-8^T showed the highest sequence similarity to *Lysobacter maris* KMU-14^T (98.6%). On phylogenetic trees, strain SG-8^T formed a stable cluster with *Lysobacter maris* KMU-14^T, *Lysobacter alkalisoli* SJ-36^T, *Lysobacter spongiae* 119BY6-57^T and *Lysobacter aestuarii* S2-C^T. The average nucleotide identity and digital DNA–DNA hybridization values between strain SG-8^T and the four reference type strains listed above were 83.3, 82.3, 83.5, 83.3% and 22.8, 22.7, 22.7, 22.9%, respectively. The major fatty acids (>5%) were iso-$C_{15:0}$, summed feature 9 (iso-$C_{17:1}$ $\omega 9c$ and/or 10-methyl $C_{16:0}$), iso-$C_{16:0}$, summed feature 3 ($C_{16:1}$ $\omega 6c$ and/or $C_{16:1}$ $\omega 7c$), iso-$C_{17:0}$, iso-$C_{11:0}$ 3OH and iso-$C_{11:0}$. Ubiquinone-8 (Q-8) was the only respiratory quinone. The major polar lipids were phosphatidylethanolamine, diphosphatidylglycerol and phosphatidylglycerol. The DNA G+C content was 68.8mol%. Based on the results of genomic, phylogenetic, phenotypic and chemotaxonomic analyses, strain SG-8^T represents a novel species of the genus *Lysobacter*, for which the name *Lysobacter penaei* sp. nov. is proposed. The type strain is SG-8^T (=GDMCC 1.1817^T=KACC 21942^T).

Strains of the genus *Lysobacter* are Gram-stain-negative, non-flagellated and flexing thin rods forming mucoid colonies [1]. Researchers are very interested in *Lysobacter* strains for their potential of producing mycolytic and bacteriolytic enzymes and antibiotic compounds [2–5]. Christensen and Cook first established the genus *Lysobacter* in 1978 [6] and, at the time of writing, this genus currently comprises 61 validly published and 12 invalidly published species (LSPN; www.bacterio.net) .

During an investigation of functional microbial strains in industrial aquaculture systems, we isolated a novel bacterial strain putatively belonging to the genus *Lysobacter*. In the present study, we aim to determine its exact taxonomic position and describe a novel *Lysobacter* species by using a polyphasic taxonomic approach.

In July 2019, a Pacific white shrimp (*Penaeus vannamei*) was collected from an indoor cement pond (750 m^2, 1.2 m depth, 21° 32′ 35″ N, 111° 22′ 38″ E) in Lingmen Town, Maoming City, Guangdong Province, PR China. The intestinal content was taken out from the shrimp and resuspended in 2 ml PBS buffer (8.0 g l^{-1} NaCl, 0.2 g l^{-1} KCl, 1.44 g l^{-1} Na_2HPO_4, 0.24 g l^{-1} KH_2PO_4; pH 7.4). After serially dilutions, 100 µl of the 10^{-6} dilution was spread onto a plate of marine agar 2216 (MA; BD Difco). After incubation at 30 °C for 48 h, a colony was picked out, purified by repeated streaking and designated as strain SG-8^T. Routinely, strain SG-8^T was preserved at −80 °C in marine broth 2216 (MB; BD Difco) supplemented with 20% glycerol, and also at 4 °C as lyophilized powder in airtight ampoules.

Author affiliations: [1]Guangdong Microbial Culture Collection Center (GDMCC), Guangdong Provincial Key Laboratory of Microbial Culture Collection and Application, State Key Laboratory of Applied Microbiology Southern China, Guangdong Institute of Microbiology, Guangdong Academy of Sciences, Guangzhou, Guangdong 510070, PR China; [2]College of Horticulture, South China Agricultural University, Guangdong Province Key Laboratory of Microbial Signals and Disease Control, Guangdong Engineering Research Center for Grass Science, Guangdong Engineering Center for Litchi, Guangzhou, 510642, PR China.
***Correspondence:** Honghui Zhu, zhuhh@gdim.cn
Keywords: *Lysobacter*; Lysobacteraceae; Lysobacterales; *Penaeus vannamei*; Pacific white shrimp; taxonomy.
Abbreviations: ANI, average nucleotide identity; dDDH, digital DNA–DNA hybridization; MA, marine agar 2216; MB, marine broth 2216.
The 16S rRNA gene and draft genome sequences of *Lysobacter penaei* SG-8^T have been deposited into GenBank under the accession numbers MT821233 and JACHTE000000000, respectively. The draft genome of *Lysobacter spongiae* LMG 30077^T has also been deposited into the GenBank database with the accession number of JACHTF000000000.
†These authors contributed equally to this work
Two supplementary figures are available with the online version of this article.

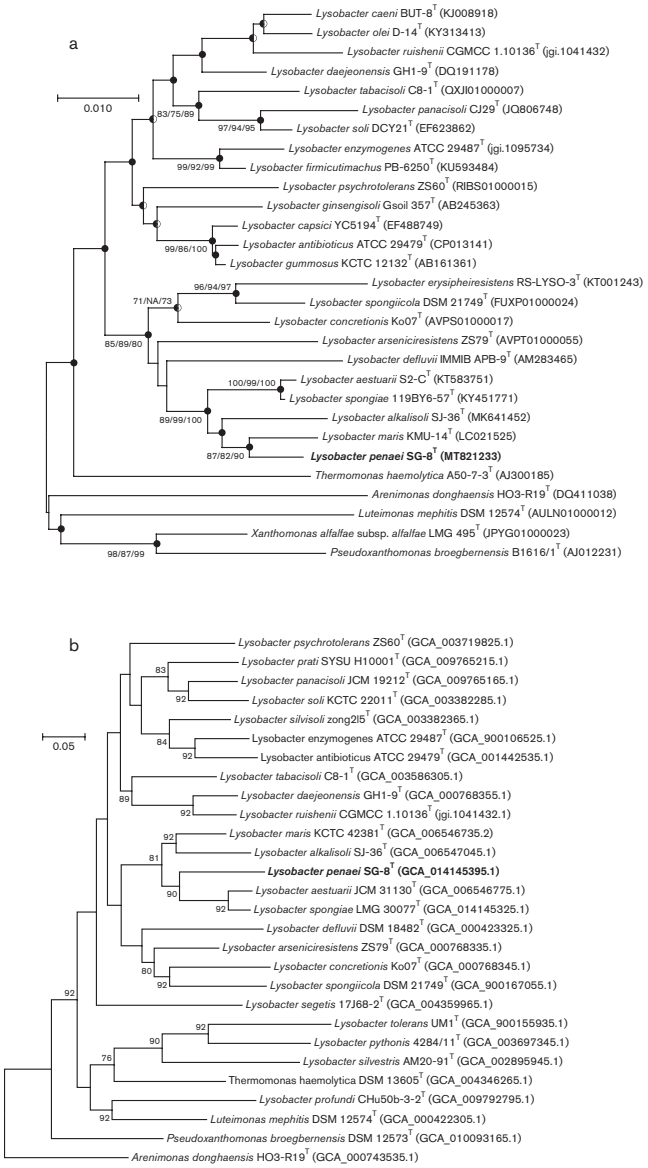

Fig. 1. (a) Neighbour-joining tree based on 16S rRNA gene sequences; (b) phylogenomic tree based on genomes. Phylogenetic trees reconstructed by the maximum likelihood (ML) and minimum evolution (ME) algorithms were mapped onto the NJ tree. ● and ◐ indicate corresponding nodes revealed in all three trees and in two of the three trees, respectively. Numbers at nodes in a and b indicated the bootstrap values based on 1000 resamplings and gene support index (GSI) values, respectively. Only values ≥70% were shown. Bar, 0.01 substitutions per nucleotide position.

The genomic DNA of strain SG-8^T was prepared by an improved CTAB method [7]. The 16S rRNA gene was amplified using the genomic DNA as PCR template and with the primers of 27F and 1492R. After sequencing by Tianyi Huiyuan Co., Ltd (Beijing, PR China), the obtained sequence was 1467 bp long and used as a query to perform similarity search on the EzBioCloud server (www.ezbiocloud.net). The 16S rRNA gene of strain SG-8^T showed the highest sequence similarity to *Lysobacter maris* KMU-14^T (98.6%), followed by *Lysobacter spongiae* 119BY6-57^T (98.1%). All other type strains in the EzBioCloud database shared 16S rRNA gene sequence similarities lower than 98.0% with strain SG-8^T. Considering that the 16S rRNA gene sequence similarities to closely related type strains were lower than the suggested threshold for prokaryotic species classification (98.65%) [8], strain SG-8^T was preliminarily regarded as a novel species and subjected to further taxonomic studies.

DNA fragmentation, library construction and genome sequencing were conducted by Shanghai Personal Biotechnology, Co., Ltd (Shanghai, PR China), on the Illumina NovaSeq platform. A total of 11204666 pair-ended raw reads and 1691904566 bp were obtained, and subsequently assembled using SPAdes version 3.11.1 with a k-mer value of 127 under the '--careful' mode [9]. After removing contigs shorter than 500 bp or with a coverage value less than 10, the genome of strain SG-8^T contained 16 contigs and 3165626 bp in total, with a coverage value of 534 and an N50 length of 346046 bp. The genomic DNA G+C content of strain SG-8^T was 68.8 mol%.

Phylogenetic analysis was performed based on 16S rRNA gene sequences and genomic sequences. The full-length 16S rRNA gene (1545 bp) obtained from the draft genome of strain SG-8^T was used to reconstruct phylogenetic trees together with 16S rRNA gene sequences of closely related type strains using MEGA-X [10]. A phylogenomic tree was also reconstructed using the pipeline of Up-to-date Bacterial Core Gene (UBCG; https://help.ezbiocloud.net/ubcg-gene-set/) [11]. Strain SG-8^T formed a stable cluster with *L. maris* KMU-14^T, *L. alkalisoli* SJ-36^T, *L. spongiae* 119BY6-57^T and *L. aestuarii* S2-C^T both in the 16S rRNA gene trees and the phylogenomic tree, suggesting that strain SG-8^T belongs to the genus *Lysobacter* (Fig. 1). For further phenotypic and chemotaxonomic characterization, *L. maris* NBRC 110750^T, *L. alkalisoli* CGMCC 1.16756^T, *L. spongiae* LMG 30077^T and *L. aestuarii* KACC 18502^T were purchased from their corresponding culture collection centres and used as reference strains.

The average nucleotide identity (ANI) and digital DNA–DNA hybridization (dDDH) values were calculated by using the ANI Calculator in EzBioCloud (www.ezbiocloud.net/tools/ani) and the Genome-to-Genome Distance Calculator (http://ggdc.dsmz.de), respectively. The ANI and dDDH values between strain SG-8^T and *L. maris* KMU-14^T (CP029843), *L. alkalisoli* SJ-36^T (CP041242), *L. spongiae* LMG 30077^T (sequenced in this study) and *L. aestuarii* S2-C^T (VICE01000000) were 83.3, 82.3, 83.5, 83.3% and 22.8, 22.7, 22.7, 22.9%, respectively, lower than the ANI threshold of 95–96% and dDDH threshold of 70% for prokaryotic species delineation.

For morphology characterization, strain SG-8^T was cultivated on MA at 30 °C for 48 h. Colonies were observed and measured by a stereoscopic microscope (SZX10, Olympus). Cells were resuspended in sterile ddH_2O and observed using transmission electron microscopy (H7650, Hitachi). Physiological and

biochemical experiments of strain SG-8^T was performed in parallel with the four reference strains under the same conditions. Gram staining, anaerobic growth, oxidase and catalase activity, hydrolysis of casein, starch, L-tyrosine, crystalline cellulose, Tweens 20, 40 and 80, growth tests under different pH, temperatures and NaCl concentrations, gliding motility and production of flexirubin-type pigments were tested as described previously [12, 13]. Also, cellular pigment was extracted with acetone and identified by the method of measuring absorption spectrum (PerkinElmer Lambda45). API 20E, 20NE and ZYM strips (bioMérieux) and Biolog GEN III MicroPlates were used to study the configured physiological characteristics, according to the manufacturer's instructions. The detailed phenotypes of strain SG-8^T are shown in Table 1,

Table 1. Phenotypic characteristics distinguishing strain SG-8^T from four closely related species of the genus *Lysobacter*

Strains: 1, SG-8^T; 2, *Lysobacter maris* NBRC 110750^T; 3, *Lysobacter spongiae* LMG 30077^T; 4, *Lysobacter aestuarii* KACC 18502^T; 5, *Lysobacter alkalisoli* CGMCC 1.16756^T; 6, *Lysobacter enzymogenes* ATCC 29487^T. +, Positive; –, negative; w, weakly positive. All strains are positive for catalase, alkaline phosphatase, esterase (C4), esterase lipase (C8), leucine arylamidase, α-chymotrypsin, acid phosphatase, naphthol-AS-Bl-phosphoamidase and hydrolysis of gelatin and casein, and negative for cystine arylamidase, β-glucuronidase, β-glucosidase, α-mannosidase, α-fucosidase, urease, fermentation of D-glucose and production of indole. All data were obtained from this study, unless indicated otherwise.

Characteristics	**1**	**2**	**3**	**4**	**5**	**6†**
Gliding motility	–	–	–	–	–	+
NaCl range for growth (%)	0–6	0–7	0–7	0–6	0–6	0–1
Tween 20	+	–	–	–	+	+
Tween 80	–	+	–	–	+	+
Starch	–	–	–	–	+	–
Tyrosine	w	w	+	+	–	+
Oxidase	+	–	+	+	+	+
API ZYM assay:						
Lipase (C14)	–	–	w	+	–	w
Valine arylamidase	w	–	w	–	–	–
Trypsin	–	w	w	w	w	+
α-Galactosidase	–	–	–	–	+	–
α-Glucosidase	–	+	–	w	–	–
N-Acetyl-β-glucosaminidase	–	+	–	+	–	+
API 20NE assay:						
Reduction of nitrate to nitrite	+	–	–	–	+	–
Denitrification	–	–	–	–	+	–
β-Galactosidase	w	w	–	–	w	–
D-Glucose	+	–	+	+	+	–
L-Arabinose	–	–	–	–	+	w
D-Mannose	–	+	+	–	+	w
D-mannitol	–	–	–	–	–	w
N-Acetyl-glucosamine	+	+	+	+	+	–
Maltose	+	+	+	+	+	–
DNA G+C content (mol%)	68.8	69.0*	69.3	69.4*	66.6*	69.4*
Genome size (Mb)	3.2	4.0*	3.6	3.5*	3.9*	6.3*
Genomic genes	2872	3578*	3417	3283*	3503*	5526*

*Calculated according to genomes downloaded from GenBank database.
†Data from Jeong *et al.* [18].

Fig. S1 (available in the online version of this article) and the species description. The characteristics listed in Table 1 clearly differentiate strain SG-8^T from the four reference strains as well as the type species of the genus *Lysobacter*.

Chemotaxonomic characterization was performed using logarithmically growing cells cultivated in MB at 30 °C for 2 days. Cellular fatty acids, polar lipids and respiratory lipoquinones were extracted and determined as described previously [14]. The major components of the cellular fatty acids of strain SG-8^T included iso-$C_{15:0}$ (20.6%), summed feature 9 (iso-$C_{17:1}$ $\omega 9c$ and/or 10-methyl $C_{16:0}$, 20.2%), iso-$C_{16:0}$ (19.3%), summed feature 3 ($C_{16:1}$ $\omega 6c$ and/or $C_{16:1}$ $\omega 7c$; 8.0%), iso-$C_{17:0}$ (7.8%), iso-$C_{11:0}$ 3OH (6.6%) and iso-$C_{11:0}$ (5.8%). According to Table 2, the fatty acid profile of strain SG-8^T was similar to those of the four reference strains and also other *Lysobacter* species [15–17], indicating that strain SG-8^T belongs to the genus *Lysobacter*. Nevertheless, some fatty acid constituents of strain SG-8^T showed differences in specific proportions, such as less iso-$C_{16:0}$ and more iso-$C_{17:0}$, summed feature 3 and summed feature 9, under synchronous experimental conditions. Strain SG-8^T contained phosphatidylethanolamine, diphosphatidylglycerol, phosphatidylglycerol, an unidentified lipid, two unidentified aminophospholipids and an unidentified aminolipid (Fig. S2). The characteristic of having phosphatidylethanolamine, diphosphatidylglycerol and phosphatidylglycerol as major polar lipids was common in strain SG-8^T and its closely related type strains [18–20]. The only respiratory quinone detected in strain SG-8^T was ubiquinone-8 (Q-8), in accordance with the fact that Q-8 is the predominant quinone type in *Lysobacter* strains [18–21].

In summary, based on the results of phylogenetic, phenotypic and physiological analyses, especially the low ANI and dDDH values with nearest neighbours, smaller genome size, fewer annotated genes, and different proportions of major fatty acids, strain SG-8^T represents a novel species in the genus *Lysobacter*, for which the name *Lysobacter penaei* sp. nov. is proposed.

Table 2. The fatty acid profiles of strain SG-8^T and four closely related type strains

Strains: 1, SG-8^T; 2, *Lysobacter maris* NBRC 110750^T; 3, *Lysobacter spongiae* LMG 30077^T; 4, *Lysobacter aestuarii* KACC 18502^T; 5, *Lysobacter alkalisoli* CGMCC 1.16756^T. All data are from this study under the same conditions. Values are percentages. Fatty acids less than 0.5% in all five strains were not shown. TR, traces (<0.5%); ND, not detected. Major components (>5%) are in bold.

Fatty acid	1	2	3	4	5
$C_{16:0}$	4.2	2.1	1.7	1.8	1.5
$C_{17:0}$ cyclo	TR	1.5	TR	TR	1.0
iso-$C_{11:0}$	**5.8**	**6.5**	**6.4**	4.9	**5.7**
iso-$C_{12:0}$	TR	TR	0.8	0.8	0.7
iso-$C_{14:0}$	1.8	1.1	3.3	3.6	1.3
iso-$C_{15:0}$	**20.6**	**20.0**	**16.8**	**15.5**	**26.3**
iso-$C_{16:0}$	**19.3**	**28.4**	**29.0**	**32.9**	**28.2**
iso-$C_{17:0}$	**7.8**	**5.1**	3.6	3.1	4.3
iso-$C_{18:0}$	0.6	TR	0.7	1.2	TR
iso-$C_{11:0}$ 3OH	**6.6**	**7.1**	**8.1**	**6.3**	4.7
iso-$C_{12:0}$ 3OH	TR	TR	TR	TR	1.7
iso-$C_{13:0}$ 3OH	ND	ND	ND	ND	0.9
iso-$C_{16:1}$ h	0.5	0.7	2.5	2.4	1.1
anteiso-$C_{15:0}$	TR	0.8	0.5	0.7	TR
anteiso-$C_{17:0}$	TR	0.7	TR	TR	TR
Summed feature 1*	TR	TR	TR	TR	0.9
Summed feature 3*	**8.0**	4.3	**5.1**	**6.5**	4.3
Summed feature 8*	1.5	1.0	1.2	1.4	0.8
Summed feature 9*	**20.2**	**16.0**	**17.0**	**15.8**	**12.1**

*As indicated by Montero-Calasanz *et al.* [22], summed features are groups of two or three fatty acids that are treated together for the purpose of evaluation in the MIDI system and include both peaks with discrete ECLs as well as those where the ECLs are not reported separately. Summer feature 1 comprises $C_{15:1}$ iso H and/or $C_{13:0}$ 3OH. Summed feature 3 comprises $C_{16:1}$ $\omega 6c$ and/or $C_{16:1}$ $\omega 7c$. Summed feature 8 comprises $C_{18:1}$ $\omega 6c$ and/or $C_{18:1}$ $\omega 7c$. Summed feature 9 comprises iso-$C_{17:1}$ $\omega 9c$ and/or 10-methyl $C_{16:0}$.

DESCRIPTION OF *LYSOBACTER PENAEI* SP. NOV.

Lysobacter penaei (pe.nae'i N.L. gen. masc. n. *penaei*, of *Penaeus*, from whose intestinal content the type strain was isolated).

After culture on MA plate at 30 °C for 48 h, colonies are light yellow, circular, smooth, shiny, creamy, convex, with entire and regular margins, and approximately 0.5 mm in diameter. Cells are Gram-stain-negative, aerobic, non-flagellated, non-gliding rods, 0.4–0.6 µm wide and 1.4–2.0 µm long. Growth occurs at 10–45 °C (optimum, 20–30 °C), pH 5.0–10.0 (optimum, pH 6.0–7.0) and in 0–6.0% (w/v) NaCl concentration (optimum, 0–4.0%). Positive for catalase, oxidase, hydrolysis of Tween 20, casein and tyrosine; negative for hydrolysis of Tweens 40 and 80, starch and cellulose. Flexirubin-type pigments are absent. Absorbance peaks of acetone-extracted pigment are at 426 nm and 449 nm, identified as α-carotenoid. In API ZYM strips, positive for alkaline phosphatase, esterase (C4), esterase lipase (C8), leucine arylamidase, valine arylamidase, α-chymotrypsin, acid phosphatase, naphthol-AS-Bl-phosphoamidase and β-galactosidase; negative for lipase (C14), cystine arylamidase, trypsin, α-galactosidase, β-glucuronidase, α-glucosidase, β-glucosidase, *N*-acetyl-β-glucosaminidase, α-mannosidase and α-fucosidase. In API 20E and 20NE strips, positive for gelatinase, reduction of nitrate, hydrolysis of aesculin, gelatin, utilization of D-glucose, *N*-acetyl-glucosamine and maltose; negative for β-galactosidase, arginine dihydrolase, lysine decarboxylase, ornithine decarboxylase, urease,

tryptophan deaminase, Voges–Proskauer test, reduction of nitrite, production of H_2S and indole, acid production from glucose, mannitol, inositol, sorbitol, rhamnose, sucrose, melibiose, amygdalin and arabinose, assimilation of L-arabinose, D-mannose, D-mannitol, potassium gluconate, capric acid, adipic acid, malic acid, trisodium citrate and phenylacetic acid. On Biolog GEN III MicroPlates, positive for utilization of dextrin, maltose, cellobiose, gentiobiose, turanose, *N*-acetyl-D-glucosamine, *N*-acetyl-β-D-mannosamine, α-D-glucose, D-fructose, D-galactose, 3-methyl glucose, D-fucose, L-fucose, L-rhamnose, D-glucose-6-phosphate, D-fructose-6-phosphate, D-aspartic acid, gelatin, glycyl-L-proline, L-alanine, L-arginine, L-aspartic acid, L-glutamic acid, L-serine, pectin, D-galacturonic acid, L-galactonic acid lactone, D-glucuronic acid, glucuronamide, mucic acid, methyl pyruvate, L-lactic acid, citric acid, Tween 40, β-hydroxy-D,L-butyric acid, α-keto-butyric acid, acetoacetic acid, propionic acid and acetic acid; negative for utilization of D-gluconic acid, sucrose, trehalose, stachyose, raffinose, lactose, melibiose, methyl β-D-glucoside, D-salicin, *N*-acetyl-D-galactosamine, *N*-acetyl-neuraminic acid, D-mannose, inosine, D-sorbitol, D-mannitol, D-arabitol, m-inositol, glycerol, D-serine, L-histidine, L-pyroglutamic acid, quinic acid, D-saccharic acid, p-hydroxy-phenylacetic acid, D-lactic acid methyl ester, α-keto glutaric acid, D-malic acid, L-malic acid, bromosuccinic acid, γ-amino butryric acid, α-hydroxy-butyric acid and formic acid. The major fatty acids (>5%) are iso-$C_{15:0}$, summed feature 9 (iso-$C_{17:1}$ $\omega 9c$ and/or 10-methyl $C_{16:0}$), iso-$C_{16:0}$, summed feature 3 ($C_{16:1}$ $\omega 6c$ and/or $C_{16:1}$ $\omega 7c$), iso-$C_{17:0}$, iso-$C_{11:0}$ 3OH and iso-$C_{11:0}$. Ubiquinone-8 (Q-8) is the only respiratory quinone. The major polar lipids are phosphatidylethanolamine, diphosphatidylglycerol and phosphatidylglycerol.

The type strain, SG-8^T (=GDMCC 1.1817^T=KACC 21942^T), was isolated from intestinal content of a Pacific white shrimp (*Penaeus vannamei*) cultured in an indoor cement pond. The DNA G+C content of the type strain is 68.8mol%. The accession numbers of the 16S rRNA gene sequence and draft genome of strain SG-8^T in GenBank are MT821233 and JACHTE000000000, respectively.

Funding information
This work was financially supported by the Key-Area Research and Development Program of Guangdong Province (2018B020205002), the National Natural Science Foundation of China (32070115), GDAS' Special Project of Science and Technology Development (2020GDASYL-20200302002), the Pearl River S and T Nova Program of Guangzhou (201806010065) and the Science and Technology Plan Project of Guangdong (2019B030316010).

Conflicts of interest
The authors declare that there are no conflicts of interest.

References

1. Brenner DJ, Krieg NR, Staley JT. Bergey's Manual of Systematic Bacteriology: Volume Two: The Proteobacteria. *Part B: The Gammaproteobacteria: Springer Science & Business Media*; 2007.
2. Li K, Hou R, Xu H, Wu G, Qian G *et al*. Two functional fatty acyl coenzyme A ligases affect free fatty acid metabolism to block biosynthesis of an antifungal antibiotic in *Lysobacter* enzymogenes. *Appl Environ Microbiol* 2020;86.
3. Afoshin AS, Kudryakova IV, Borovikova AO, Suzina NE, Toropygin IY *et al*. Lytic potential of *Lysobacter capsici* VKM B-2533T: bacteriolytic enzymes and outer membrane vesicles. *Sci Rep* 2020;10:13.
4. LJ Y, Khetrapal V, Liu FQ, LC D. LeTetR positively regulates 3-hydroxylation of the antifungal HSAF and its analogs in *Lysobacter* enzymogenes OH11. *Molecules* 2020;25:10.
5. Cimmino A, Bejarano A, Masi M, Puopolo G, Evidente A. Isolation of 2,5-diketopiperazines from *Lysobacter capsici* AZ78 with activity against *Rhodococcus fascians*. *Nat Prod Res* 2020;24:1–9.
6. Christensen P, Cook F. Lysobacter, a new genus of nonfruiting, gliding bacteria with a high base ratio. *International Journal of Systematic and Evolutionary Microbiology* 1978;28:367–393.
7. Chen WP, Kuo TT. A simple and rapid method for the preparation of gram-negative bacterial genomic DNA. *Nucleic Acids Res* 1993;21:2260.
8. Kim M, Oh H-S, Park S-C, Chun J. Towards a taxonomic coherence between average nucleotide identity and 16S rRNA gene sequence similarity for species demarcation of prokaryotes. *Int J Syst Evol Microbiol* 2014;64:346–351.
9. Bankevich A, Nurk S, Antipov D, Gurevich AA, Dvorkin M *et al*. SPAdes: a new genome assembly algorithm and its applications to single-cell sequencing. *J Comput Biol* 2012;19:455–477.
10. Kumar S, Stecher G, Li M, Knyaz C, Tamura K. MEGA X: molecular evolutionary genetics analysis across computing platforms. *Mol Biol Evol* 2018;35:1547–1549.
11. SI N, Kim YO, Yoon SH, SM H, Baek I *et al*. UBCG: up-to-date bacterial core gene set and pipeline for phylogenomic tree reconstruction. *J Microbiol* 2018;56:280–285.
12. Li A, Zhang M, Xu S, Chen M, Yao Q *et al*. *Pseudidiomarina gelatinasegens* sp. nov., isolated from surface sediment of the Terra Nova Bay, Antarctica. *Int J Syst Evol Microbiol* 2020;70:708–714.
13. Li A-Z, Lin L-Z, Zhang M-X, Zhu H-H. *Arenibacter antarcticus* sp. nov., isolated from marine sediment. *Int J Syst Evol Microbiol* 2017;67:4601–4605.
14. Li A-Z, Lin L-Z, Zhang M-X, Lv Y, Zhu H-H. *Arenibacter catalasegens* sp. nov., isolated from marine surface sediment, and emended description of the genus *Arenibacter*. *Int J Syst Evol Microbiol* 2018;68:758–763.
15. Zhang L, Bai J, Wang Y, Wu G-L, Dai J *et al*. *Lysobacter korlensis* sp. nov. and *Lysobacter bugurensis* sp. nov., isolated from soil. *Int J Syst Evol Microbiol* 2011;61:2259–2265.
16. Jin C-Z, Song X, Sung YJ, Jin F-J, Li T *et al*. *Lysobacter profundi* sp. nov., isolated from freshwater sediment and reclassification of *Lysobacter panaciterrae* as *Luteimonas panaciterrae* comb. nov. *Int J Syst Evol Microbiol* 2020;70:3878–3887.
17. Lee D, Jang JH, Cha S, Seo T. *Lysobacter humi* sp. nov., isolated from soil. *Int J Syst Evol Microbiol* 2017;67:951–955.
18. Jeong SE, Lee HJ, Jeon CO. *Lysobacter aestuarii* sp. nov., isolated from estuary sediment. *Int J Syst Evol Microbiol* 2016;66:1346–1351.
19. Xu L, Huang X-X, Fan D-L, Sun J-Q. *Lysobacter alkalisoli* sp. nov., a chitin-degrading strain isolated from saline-alkaline soil. *Int J Syst Evol Microbiol* 2020;70:1273–1281.
20. Choi H, Im W-T, Park J-S. *Lysobacter spongiae* sp. nov., isolated from spongin. *Journal of Microbiology* 2018;56:97–103.
21. Yoon J. Polyphasic characterization of *Lysobacter maris* sp. nov., a bacterium isolated from seawater. *Current Microbiology* 2016;72:282–287.
22. Montero-Calasanz MdelC, Göker M, Rohde M, Spröer C, Schumann P *et al*. *Chryseobacterium hispalense* sp. nov., a plant-growth-promoting bacterium isolated from a rainwater pond in an olive plant nursery, and emended descriptions of *Chryseobacterium defluvii*, *Chryseobacterium indologenes*, *Chryseobacterium wanjuense* and *Chryseobacterium gregarium*. *Int J Syst Evol Microbiol* 2013;63:4386–4395.