Amphiplicatus metriothermophilus gen. nov., sp. nov., a thermotolerant alphaproteobacterium isolated from a hot spring

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A thermotolerant, Gram-strain-negative, non-spore-forming and strictly aerobic bacterium, designated GU51^T, was isolated from Guhai hot spring in Jimsar county, Xinjiang province, north-west China. Each cell of strain GU51^T consisted of an oval body and two symmetrical long (3–6 μm) prosthecae. The strain moved by polar flagellum. Oxidase and catalase were produced. Strain GU51^T grew within the ranges of 37–65 °C (optimum 48–50 °C), 0.5–7.5 % (w/v) NaCl (optimum 2–3 %) and pH 6.0–9.0 (optimum pH 7.5). The major respiratory quinone detected was ubiquinone 10 (U-10) and the genomic DNA G+C content was 66.7 ± 0.4 mol%. Major fatty acids (>5 %) were $C_{16:0}$, $C_{18:1}\omega7c$ and 11-methyl $C_{18:1}\omega7c$. The polar lipids consisted of diphosphatidylglycerol, five glycolipids, phosphatidylglycerol and an unknown phospholipid. Phylogenetic analysis showed the closest relatives of strain GU51^T were members of the genus *Parvularcula* with 92.3 % 16S rRNA gene sequence similarity. On the basis of this polyphasic taxonomic characterization, it is suggested that strain GU51^T represents a novel species of a new genus in the family '*Parvularculaceae*', for which the name *Amphiplicatus metriothermophilus* gen. nov., sp. nov. is proposed. The type strain of the type species is GU51^T (=CGMCC 1.12710^T=JCM 19779^T).

The family '*Parvularculaceae*' belongs to the order '*Parvularculales*'. At the time of writing, the order comprises one family and the family comprises a single genus, *Parvularcula*, which was established by Cho & Giovannoni (2003) and consists of three species with validly published names: *Parvularcula bermudensis* (Cho & Giovannoni, 2003), *Parvularcula lutaonensis* (Arun *et al.*, 2009) and *Parvularcula dongshanensis* (Yu *et al.*, 2013). The isolates belonging to the genus *Parvularcula* were obtained from seawater, a coastal hot spring and a soft coral, respectively, which indicates that the evolutionary cluster had a high similarity of habitat and metabolism pathways. All three species of the genus are mesophilic, Gram-strain-negative and non-spore-forming. Phylogenetic tree analysis revealed that a novel thermotolerant strain, designated GU51^T, was closely related to the genus *Parvularcula*. However, it was clearly distinct from this genus by high sequence divergence values and the significant differences of morphological and chemotaxonomic characteristics. In the present study, we report the results of a polyphasic taxonomy on strain GU51^T and propose that strain GU51^T represents a novel species of a new genus in the family '*Parvularculaceae*'.

Strain GU51^T was isolated from the Guhai hot spring (44° 45′ N 88° 49′ E) in Jimsar county, Xinjiang province, north-west China. The highest temperature *in situ* was measured to be 75 °C. Guhai hot spring, which means ancient sea, was discovered accidentally during oil exploration in October1982 (official reports). A water sample from the spring was collected and transported without temperature control and then stored at room temperature in the laboratory until used. 0.52 g crystals were produced from evaporation of 20 ml water sample at 55 °C in the laboratory, and were sent for detection. According to the analysis results of X-ray Energy Dispersive Spectroscopy (EDS) (GENESIS4000; EDAX), several elements such as O, Na, S, Cl and Ca were at high levels of 16.22 %, 10.95 %,

Abbreviations: DPG, diphosphatidylglycerol; PG, phosphatidylglycerol.

The GenBank/EMBL/DDBJ accession number for the 16S rRNA gene sequence of strain GU51^T is KF153051.

Four supplementary figures are available with the online version of this paper.

1.19 %, 49.87 % and 21.77 %, respectively. To obtain the enrichment culture, 400 ml spring water was filtered (0.22 μm, SCRC) and the membrane was subsequently inoculated into modified marine 2216 medium, the composition of which was the same as marine broth 2216 (BD) except that 5 g l^{-1} trypticase peptone (BD) and 0.01 g l^{-1} ferric citrate were added. Parallel preparations were cultivated at 50 °C and 120 r.p.m. in a rotary water-bath shaker. The turbid cultures were serially diluted and spread onto modified marine 2216 agar. A single colony was picked out, purified at least three times before cultivating in modified marine 2216 medium for 3 days at 50 °C and storing at −80 °C with 30 % (v/v) glycerol. Reference strains including *Thermovum composti* KCTC 23707^T (Yabe *et al.*, 2012) which has the highest 16S rRNA gene sequence similarity value with strain GU51^T, and *Parvularcula bermudensis* KCTC 12087^T which was located closest to strain GU51^T in the phylogenetic tree, were purchased from the Korean Collection for Type Cultures.

The temperature, pH and NaCl ranges for growth were determined by measuring the OD_{600} every six hours. The temperature was respectively tested at 28, 35, 37, 43, 45, 48, 50, 53, 55, 60 and 65 °C. For salt tolerance, 0, 1, 2, 3, 5, 7 and 10 % NaCl (w/v) were added to NaCl-free modified 2216 medium. The pH range for growth was determined using different buffering agents including MES (for pH 5.5–6.5), MOPS (pH 6.5–7.5), Tris (pH 7.5–8.5) and Bis (pH 8.0–9.5). Anaerobic growth was detected by using the Hungate roll-tube technique (Hungate, 1969) with modified marine 2216 agar. Cultures were incubated for over 15 days under pure N_2.

Unless otherwise mentioned, strain GU51^T and the reference strains were all cultivated at the respective optimal growth conditions. Cell morphology was examined by optical microscopy (BX40; Olympus) and transmission electron microscopy (JEM-1230; JEOL) using cultures incubated for 30 h (adjustment phase), 70 h (exponential phase), 84 h (stationary phase) and 115 h (decline phase).

Catalase and oxidase activities were tested as described by Zhang *et al.* (2013). H_2S production was tested as described by Shen & Chen (2008). Selenite reduction was performed using the method described by Mata *et al.* (2002). Alginate lyase was tested by spreading 70 % ethanol onto algin plates (1 %, w/v) (Kawasaki *et al.*, 2002). Hydrolysis of CM-cellulose (0.2 %, w/v), pectin (0.2 %, w/v) and xylan (0.2 %, w/v) were tested by flooding the corresponding plates with Congo red reagent (0.2 %, w/v). Nitrate and nitrite reduction, hydrolysis of starch (2 %, w/v), casein (1 %, w/v), skimmed milk (1 %, w/v), Tweens 20, 40, 60 and 80 (0.5 %, v/v), aesculin (0.1 %, w/v), gelatin (1 %, w/v) and DNA (0.4 %, w/v) were tested according to Dong & Cai (2001). All experiments were performed in triplicate. Other tests were carried out using API 20 NE and API ZYM strips (bioMérieux) and GN2 MicroPlates (Biolog) following the manufacturers' instructions. Original media (AUX medium) and artificial seawater were mixed at equal volumes and the mixture was used as the basal medium to suspend cells for the GN2 MicroPlate test. Artificial seawater contained (per litre distilled water): 20 g NaCl, 5 g $MnCl_2.2H_2O$, 2 g $MnSO_4.7H_2O$, 0.5 g $CaCl_2$ and 1 g KCl. The results are provided in the species description. Acid production was tested using modified marine 2216 medium supplemented with 0.5 % (w/v) carbohydrates and 2.5 % (w/v) bromocresol purple (Liu *et al.*, 2014).

Sensitivity to antibiotics was assayed with a two-layer plate method as described previously (Zhang *et al.*, 2010) except that modified marine 2216 agar was used. The strains were considered susceptible when the diameter of the inhibition zone was >5 mm, intermediate at 2–5 mm and resistant at <2 mm as described by Nokhal & Schlegel (1983). Each antibiotic (Hangzhou Microbial Reagent Co.) was tested in triplicate. The results showed that strain GU51^T and the reference strains were all sensitive to carbenicillin (100 μg), chloramphenicol (30 μg), macrodantin (300 IU), neomycin (30 μg) and rifampicin (5 μg), but were resistant to naphthridinic acids (30 μg) and polymyxin B (300 IU). In addition, strain GU51^T was also sensitive to amoxicillin (10 μg), kanamycin (30 μg) and mefoxin (30 μg), and resistant to ampicillin (10 μg), cephalothin (30 μg), gentamicin (10 μg), jiemycin (2 μg), oxacillin (1 μg), streptomycin (10 μg) and tobramycin (10 μg), which was quite different from the reference strains. Detailed results are shown in Table 1.

Cells of strain GU51^T and the reference strains used for polar lipid, respiratory quinone and fatty acid analyses were obtained from cultures incubated in flasks containing 250 ml modified marine 2216 medium until exponential phase. Polar lipids were extracted as described previously (Kates, 1986). Extracts were separated by two-dimensional TLC with silica gel 60 F_{254} plates (Merck) which were then sprayed with sulfuric acid/ethanol (1 : 1, v/v) and heated at 120 °C for 10 min as specified by Cui *et al.* (2011). The LC-MS system (Agilent) was used for quinone analysis. The identification and quantification of fatty acid methyl esters were performed using the Sherlock Microbial Identification System (MIDI). Each experiment was carried out twice to confirm the results.

Genomic DNA was extracted as described by Marmur (1961). The DNA G+C content of strain GU51^T was determined by reverse-phase HPLC as described previously (Mesbah & Whitman, 1989). PCR amplification of the 16S rRNA gene was performed using bacterial universal primers 27F (5′-AGAGTTTGATCCTGGCT-3′) and 1492R (5′-GGTTACCTTGTTACGACTT-3′). PCR products were cloned into the pMD 19-T vector (TaKaRa) and then sequenced by the Sanger method using an ABI Prism 3730 DNA sequencer (Applied Biosystems) as described by Xu *et al.* (2007). The GenBank database (http://blast.ncbi.nlm.nih.gov/Blast.cgi) and the EzTaxon-e service (http://eztaxon-e.ezbiocloud.net/, Kim *et al.*, 2012) were utilized for sequence alignment. All related sequences were exported to the MEGA version 5.05 software package (Tamura *et al.*,

Table 1. Differential characteristics between strain GU51^T and reference strains

Strains: 1, GU51^T (data from this study); 2, *T. composti* KCTC 23707^T (this study); 3, *Parvularcula bermudensis* KCTC 12087^T (this study); 4, *Parvularcula lutaonensis* KCTC 22245^T (Arun *et al.*, 2009); 5, *Parvularcula dongshanensis* MCCC 1A06534^T (Yu *et al.*, 2013). Strains 1–3 were positive for oxidase activity and nitrate reduction, and negative for indole production, hydrolysis of algin, CM-cellulose, DNA, pectin, starch, xylan, casein and Tweens 40, 60 and 80. +, Positive; −, negative; W, weak; ND, not detected.

Characteristic	1	2	3	4	5
Isolation source	Hot spring water	Mature compost	Seawater	Hot spring	Soft coral
Colony colour	Light yellow	Cream	Yellowish-brown	Orange	Red
Temperature range for growth (optimum) (°C)	37–65 (48–50)	23–57 (50)	10–37 (30)	25–50 (37)	16–41 (28)
pH range for growth (optimum)	6.0–9.0 (7.5)	5.9–8.8 (7.0)	6.0–9.0 (8.0)	7–9(7.0)	ND
NaCl tolerance concentration (optimum) (%, w/v)	0.5–7.5 (2–3)	4–8 (4.5)	0.75–20 (3.0)	0.5–6 (3.0)	0–12 (1–3)
Gram stain	−	+	−	−	−
H_2S production	+	+	−	−	ND
Selenite reduction	+	−	+	ND	ND
Catalase	+	+	−	−	−
Hydrolysis of:					
Aesculin	+	−	−	+	+
Gelatin	+	−	+	+	+
Skimmed milk	−	−	+	ND	ND
Tween 20	+	−	−	+	ND
Enzyme activities (API ZYM)					
Alkaline phosphatase	+	−	+	+	+
Valine arylamidase	+	+	W	+	−
Acid phosphatase, trypsin	+	+	W	+	+
Cystine arylamidase	W	−	W	+	−
α-Chymotrypsin	+	W	W	+	W
α-Galactosidase,	W	−	−	−	+
α-Glucosidase	+	W	−	+	+
β-Glucosidase	+	−	−	−	+
N-Acetyl-β-glucosaminidase	+	−	−	−	−
Enzyme activities (API 20NE)					
Urea	−	+	+	−	−
Arginine dihydrolase	−	−	+	−	−
β-Galactosidase	+	−	−	+	+
Antibiotic resistance					
Mefoxin, amoxicillin	+	+	−	ND	ND
Kanamycin	+	−	+	ND	−
Streptomycin, gentamicin	−	−	+	ND	+
Tobramycin	−	−	+	ND	ND
Jiemycin, cephalothin	−	+	−	ND	ND
Ampicillin	−	+	−	ND	+
Oxacillin	−	+	−	−	−
Acid production from:					
Rhamnose	+	−	+	ND	ND
Xylose	−	+	−	ND	ND
Cellulose, sorbitol	−	−	+	ND	ND
Arabinose	+	+	−	ND	ND
Raffinose	+	−	−	ND	ND
DNA G+C content (mol%)	66.7±0.4	63.2±0.8	61.2±0.8	59.0±1	61.8

2011) for multiple sequence alignments and phylogenetic tree reconstruction. Phylogenetic analysis using neighbour-joining (Saitou & Nei, 1987), maximum-parsimony and maximum-likelihood (Felsenstein, 1981) methods was performed. Evolutionary distances used in the neighbour-joining and maximum-likelihood analyses were calculated according to the algorithm of Kimura's two-parameter model (Kimura, 1980).

Cells of strain GU51^T were Gram-strain-negative, non-spore-forming, motile and strictly aerobic. Strain GU51^T grew at 37–65 °C (optimum 48–50 °C), pH 6.0–9.0 (optimum 7.5) and in the presence of 0.5–7.5 % (w/v)

NaCl (optimum 2–3 %). After incubation at 50 °C for 3 days, colonies on modified marine 2216 agar were circular, elevated and light yellow with smooth edges.

The cellular morphology of strain GU51^T is shown in Fig. 1 and Fig. S1 (available in the online Supplementary Material). Based on the observation of cells obtained from different growth phases, two polar prosthecae with variable length occurred symmetrically during most of the lifetime of strain GU51^T. This morphology was quite stable until stationary phase. When growth time extended to more than 100 h (Fig. S1c), the prosthecae were becoming detached. In stationary phase, cells were 0.25–0.5 μm in diameter, 0.5–1 μm in length (Fig. 1a) and the prosthecae were about 0.075–0.1 μm wide and 3–6 μm long (Fig. 1b). The prosthecae, which were surrounded by bilayer membrane, consisted of a series of bubbles (Fig. 1b). Moreover, the thin-section electron micrograph indicated that the long prosthecae were connected with the cell membrane (Fig. S1d). Thus, it was suggested that the long prosthecae of GU51^T were formed by the extension of cell membrane. The cellular morphology of members of the genus *Parvularcula* was short rods and that of the genus *Thermovum* was oval-shaped; therefore the morphology of strain GU51^T is distinctive when compared with the shape of these two genera.

The major polar lipids of strain GU51^T consisted of diphosphatidylglycerol (DPG), phosphatidylglycerol (PG) and two glycolipids (GL) (Fig. S2). There was one more glycolipid in *Parvularcula bermudensis* KCTC 12087^T compared with strain GU51^T and the content of PG was very different between them. The unidentified phospholipid PL2 detected in *Parvularcula bermudensis* KCTC 12087^T could not be found in strain GU51^T. Meanwhile, it was only an unknown phospholipid (PL) in GU51^T which was quite different from *Parvularcula lutaonensis* KCTC22245 (four unknown phospholipids) (Arun *et al.*, 2009) and *Parvularcula dongshanensis* MCCC 1A06534 (five unknown phospholipids) (Yu *et al.*, 2013). The presence of DPG could be another evidence to differentiate strain GU51^T from the two strains.

The fatty acid profile of strain GU51^T was distinct from members of the genera *Parvularcula* and *Thermovum* (Table 2). Strain GU51^T contained $C_{16:0}$ (38.5 %) as its predominant fatty acid of which the content was only about 15 % in members of the genera *Thermovum* and *Parvularcula*. The presence of 11-methyl $C_{18:1}\omega 7c$ (8.2 %) in strain GU51^T which has not been detected in the type strains of *Parvularcula lutaonensis* and *Parvularcula dongshanensis* and the great content difference of this fatty acid in *Parvularcula bermudensis* KCTC 12087^T (0.3 %) compared to strain GU51^T could be another evidence to differentiate strain GU51^T from the reference strains.

A 16S rRNA gene sequence of 1470 nt was obtained from strain GU51^T. The result of sequence alignments showed that *Thermovum composti* KCTC 23707^T shared the highest 16S rRNA gene sequence similarity value of 92.5 % with strain GU51^T. Other type strains which showed comparatively high sequence similarities (>91 %) to strain GU51^T were *Methyloligella halotolerans* DSM 25045^T (92.2 %) (Doronina *et al.*, 2013) in the family *Hyphomicrobiaceae*; *Afifella pfennigii* DSM 17143^T (91.8 %) (Caumette *et al.*, 2007; Urdiain *et al.*, 2008) in the family *Rhodobiaceae*; *Mesorhizobium loti* JCM 21464^T (91.6 %) (Jarvis *et al.*, 1982, 1997) in the family *Phyllobacteriaceae*; *Parvularcula bermudensis* KCTC 12087^T (92.3 %) in the family '*Parvularculaceae*'; and *Phenylobacterium lituiforme* DSM 14363^T (91.3 %) (Kanso and Patel, 2004) in the family *Caulobacteraceae*.

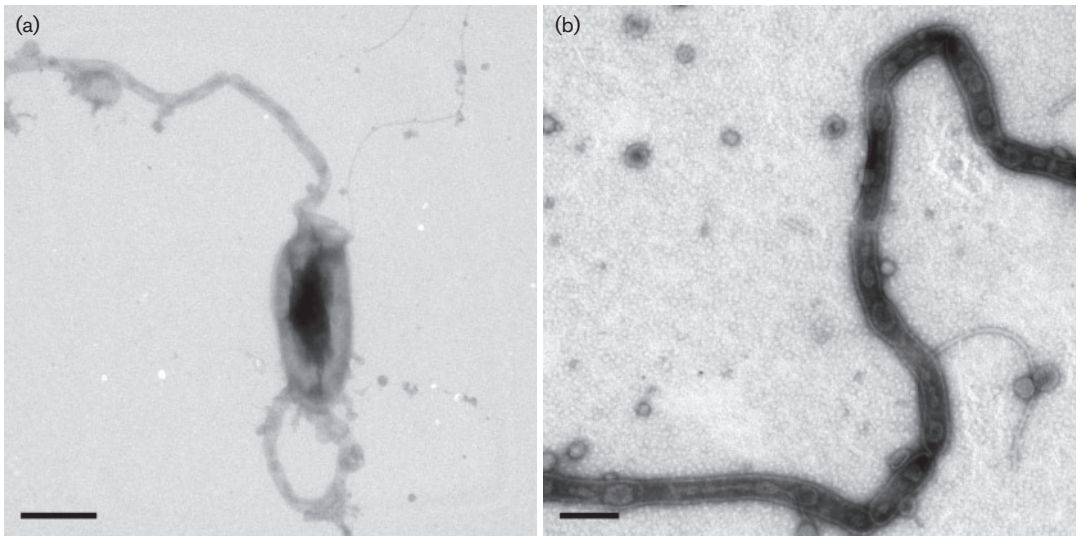

Fig. 1. Transmission electron micrographs of cells of strain GU51^T growing on modified marine 2216 agar at 50 °C. (a) Hanging-drop at stationary phase (84 h) and (b) close-up of the long prosthecae of exponential phase. Bars, 0.5 μm (a) and 0.2 μm (b).

Table 2. Fatty acid content (%) of strain GU51^T and the reference strains

Taxa: 1, strain GU51^T; 2, *T. composti* KCTC23707; 3, *Parvularcula bermudensis* KCTC12087; 4, *Parvularcula lutaonensis* KCTC 22245; 5, *Parvularcula dongshanensis* MCCC 1A06534. Data for taxa 1–3 were generated in this study; data for taxa 4–5 were taken from previous studies (Arun *et al.*, 2009; Yu *et al.*, 2013). ND, Not detected; TR, trace amount (<1.0 %).

Fatty acid	1	2	3	4	5
$C_{16:0}$	38.5	16.3	10.1	15.7	22.7
$C_{18:1}\omega7c$	32.4	11.0	67.6	73.4	ND
11-methyl $C_{18:1}\omega7c$	8.2	0.3	ND	ND	ND
$C_{19:0}$cyclo $\omega8c$	4.6	44.0	ND	ND	3.4
$C_{14:0}$ 2-OH	3.9	ND	ND	ND	9.4
Summed feature 3*	3.7	0.9	0.6	ND	3.0
$C_{18:0}$	1.6	14.4	1.3	5.7	4.0
$C_{17:0}$	1.3	6.8	0.2	ND	TR
$C_{12:0}$	1.0	ND	9.7	1.2	1.8
iso-$C_{17:0}$	1.0	ND	0.2	1.9	ND
$C_{15:0}$	0.7	0.6	0.3	ND	ND
$C_{14:0}$	0.5	0.5	5.3	ND	1.5
$C_{16:1}\omega9c$	0.4	ND	ND	ND	ND
iso-$C_{15:0}$	0.4	ND	ND	ND	ND
$C_{16:0}$ 2-OH	0.3	ND	0.1	ND	2.1
$C_{17:1}\omega6c$	0.3	ND	ND	ND	ND
$C_{15:0}$ 2-OH	0.2	ND	ND	ND	ND
$C_{17:1}\omega8c$	0.2	0.4	ND	ND	ND
iso-$C_{17:1}\omega9c$	0.2	ND	ND	ND	ND
Summed feature 8*	ND	ND	ND	ND	49.4

*Summed features represent groups of two or three fatty acids that cannot be separated by GLC with the MIDI system. Summed feature 3 comprised iso-$C_{15:0}$ 2-OH/$C_{16:1}\omega7c$; summed feature 8 comprised $C_{18:1}\omega7c/\omega6c$.

T. composti is the only species in the genus *Thermovum*. Phylogenetic analysis indicated that though the type strain of *T. composti* shared the highest 16S rRNA gene sequence similarity with strain GU51^T, it did not cluster with the novel isolate in the phylogenetic tree (Fig. 2). Instead, type strains of species of the genus *Parvularcula* showed the closest phylogenetic relationship with strain GU51^T by clustering in the same branch with a high bootstrap value. Moreover, the cluster formed by strain GU51^T and members of the genus *Parvularcula* was unattached to other clades respectively designated into families *Hyphomicrobiaceae, Rhodobiaceae, Phyllobacteriaceae* and *Caulobacteraceae*, and thus was considered to represent the branch of family '*Parvularculaceae*' to which the genus *Parvularcula* belongs. The phylogenetic topology exhibited in Fig. 2 was also supported by the maximum-likelihood (Fig. S3) and maximum-parsimony trees (Fig. S4). In conclusion, considering the high dissimilarity (>7.7 %) in 16S rRNA gene sequence and the relatively distant evolutionary distance even with its closest relatives genus *Parvularcula*, strain GU51^T was considered to represent a novel taxon of family '*Parvularculaceae*'.

Strain GU51^T could also be differentiated from the genera *Thermovum* and *Parvularcula* on the basis of several properties, including optimal temperature for growth, NaCl tolerance, H_2S production, catalase activity and hydrolysis of aesculin and Tween 20.

Therefore, taking all characteristics into consideration, strain GU51^T was clearly distinct from the genus *Parvularcula* by high sequence divergence values and the significant differences in morphological and chemotaxonomic characteristics. We propose that strain GU51^T represents a novel species of a new genus in the family '*Parvularculaceae*', for which we offer the name *Amphiplicatus metriothermophilus* gen. nov., sp. nov.

Description of *Amphiplicatus* gen. nov.

Amphiplicatus (Am.phi.pli.ca′tus. Gr. pref. *amphi* both sides or double; L. part. adj. *plicatus* folded; N.L. masc. n. *Amphiplicatus* folded on both sides, referring to the long plate-like prosthecae of the cells).

Cells are Gram-strain-negative, thermophilic and motile with a single flagellum. Strictly aerobic. No spores are observed. Catalase- and oxidase-positive. Nitrate is reduced to nitrite. The major respiratory quinone is U-10. The G+C content of the genomic DNA is 66.7±0.4 mol%. The polar lipids consist of DPG, PG, five glycolipids and an unknown phospholipid. Major fatty acids (>5 %) are $C_{16:0}$, $C_{18:1}\omega7c$ and 11-methyl $C_{18:1}\omega7c$. Phylogenetically, the genus is affiliated to the family '*Parvularculaceae*' of the order '*Parvularculales*'.

The type species is *Amphiplicatus metriothermophilus*.

Description of *Amphiplicatus metriothermophilus* sp. nov.

Amphiplicatus metriothermophilus (me.tri.o.ther.mo′phi.lus. Gr. adj. *metrios* modest; Gr. n. *therme* heat; Gr. adj. *philos* friend, loving; N.L. masc. adj. *metriothermophilus* modestly heat-loving).

Displays the following characteristics in addition to those in the genus description. Colonies on modified marine 2216 agar after 3 days of incubation at 50 °C are light yellow, smooth, circular and elevated. Grows at 37–65 °C (optimum 48–50 °C). The pH range for growth is 6.0–9.0 (optimum pH 7.5). Growth occurs in the presence of 0.5–7.5 % (w/v) NaCl (optimum 2–3 %). Degrades Tween 20, but not Tweens 40, 60 and 80, starch or CM-cellulose. Positive for aesculin hydrolysis, but negative for hydrolysis, indole production, arginine dihydrolase and urease. Cells produce H_2S and can reduce sodium hydrogen selenite to element of selenium. With GN2 MicroPlates, α-cyclodextrin, dextrin, glycogen, *N*-acetyl-D-glucosamine, adonitol, D-arabitol, lactulose, β-hydroxybutyric acid, L-ornithine and glycerol are utilized; all other carbon sources are not

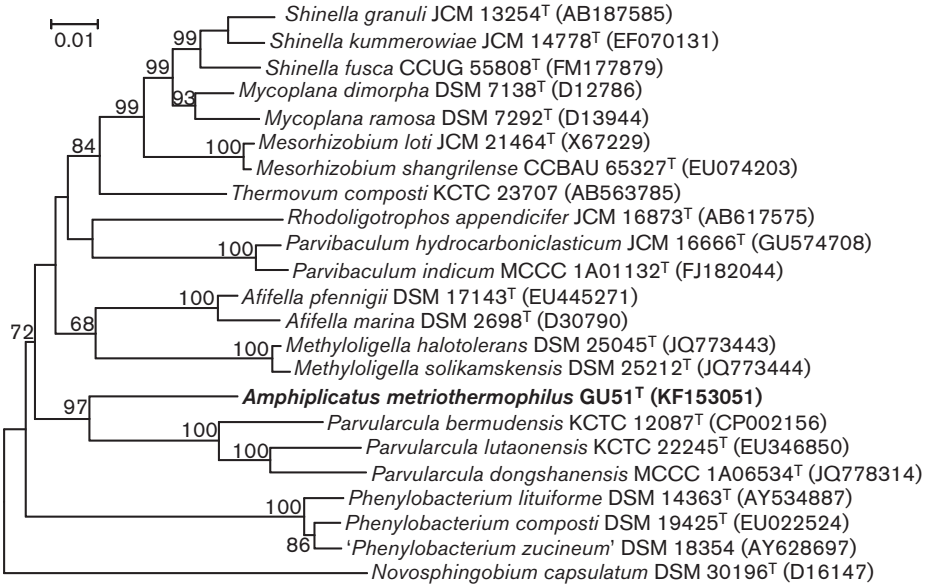

Fig. 2. Neighbour-joining tree based on the 16S rRNA gene sequences of strain GU51^T and related taxa. Numbers at nodes are bootstrap percentages based on 1000 replicated datasets. The sequence of *Novosphingobium capsulatum* DSM 30196^T was used as an outgroup. DDBJ/EMBL/GenBank accession numbers are shown in parentheses. Bar, 0.01 substitutions per nucleotide position.

utilized. In API ZYM tests, positive results for alkaline phosphatase, esterase (C4), esterase lipase (C8), leucine arylamidase, valine arylamidase, cystine arylamidase (weak), trypsin, α-chymotrypsin, acid phosphatase, naphthol-AS-BI-phosphoamidase, α-galactosidase (weak), β-galactosidase (weak), α-glucosidase, β-glucosidase and *N*-acetyl-β-glucosaminidase activities, but negative results for β-glucuronidase, lipase (C14), α-mannosidase and α-fucosidase activities.

The type strain GU51^T (=CGMCC 1.12710^T=JCM 19779^T) was isolated from a water sample of Guhai hot spring in Jimsar county, Xinjiang province, north-west China. The DNA G+C content of the type strain is 66.7±0.4 mol%.

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