Sulfurimonas aquatica sp. nov., a sulfur-oxidizing bacterium isolated from water of a brackish lake

Hisaya Kojima*, Yukako Kato, Tomohiro Watanabe, and Manabu Fukui

The Institute of Low Temperature Science, Hokkaido University. Kita-19, Nishi-8, Kita-ku, Sapporo 060-0819, Japan

*Corresponding author

Abstract

A novel chemolithoautotrophic bacterium, strain H1576^T, was isolated from water of a brackish lake. Strain H1576^T grew aerobically on inorganic sulfur compounds. Hydrogen gas did not support autotrophic growth, and heterotrophic growth was not observed. Cells were rod-shaped, motile, 1.5–2.7 μm in length and 0.6–0.7 μm in width. Growth was observed at 3–22°C with an optimum growth temperature of 13–15°C. The pH range for growth was 6.0–7.4 with an optimum pH of 6.6–6.8. Major fatty acids were summed feature 3 ($C_{16:1}\omega7c$ and/or $C_{16:1}\omega6c$). The complete genome of strain H1576^T consists of a circular chromosome and a plasmid, with total length of 2.8 Mbp and G + C content of 46.4 mol%. Phylogenetic analyses indicated that strain H1576^T belongs to the genus *Sulfurimonas* but distinct from representatives of existing species. On the basis of genomic and phenotypic characteristics, a new species named *Sulfurimonas aquatica* sp. nov. is proposed with the type strain of strain H1576^T (= BCRC 81254^T = JCM 35004^T).

Introduction

According to the List of Prokaryotic Names with Standing in Nomenclature, LPSN (Parte et al., 2020), the genus *Sulfurimonas* belongs to the family *Helicobacteraceae* and currently includes eight species with validly published names (as of 26 July 2022). They grow chemolithoautotrophically by oxidizing inorganic sulfur compounds, with oxygen as electron acceptor. In some species, anaerobic growth and H_2 gas oxidation are observed. As chemotaxonomic feature, they share major fatty acids of $C_{16:1}$, $C_{18:1}$ and $C_{16:0}$. Besides these eight species, three other species and two *Candidatus* species have been proposed in this genus, on the basis of genomic and phenotypic characterizations of isolated strains (Table 1).

As reviewed previously (Han & Perner, 2015), members of the genus *Sulfurimonas* have been repeatedly detected by 16S rRNA gene sequence analysis, in various ecosystems represented by hydrothermal vents, marine sediments and water columns. In addition, *Sulfurimonas* is known to be a dominating bacterial genus in some engineered microbial systems, as shown in recent studies employing 16S rRNA gene amplicon sequencing (Fu et al., 2020; Wu et al., 2020; Haosagul et al, 2021). With the same approach, a dominance of *Sulfurimonas* species at specific water depths of a stratified brackish lake was recently reported (Watanabe et al., 2022). This shallow

47 eutrophic lake, Lake Harutori in Japan, is characterized by steep chemocline and high
48 concentration of sulfide in bottom water (Kubo et al, 2014; Watanabe et al., 2022). In this
49 study, a novel sulfur-oxidizing autotroph was isolated from anoxic water of Lake Harutori,
50 and characterized as a representative of a new species in the genus *Sulfurimonas*.
51
52 **Materials and methods**
53 Sampling of water from Lake Harutori was conducted on 16 Feb 2016. A sample
54 of anoxic bottom water was collected from 5 m depth, at a site where previous studies
55 were conducted (Kubo et al., 2014; Watanabe et al., 2022). A portion of the sample (0.3
56 ml) was inoculated into 30 ml of a medium for aerobic thiosulfate oxidizers. The medium
57 (hereafter referred to as basal medium) was prepared as described below. First, the
58 following salts (g l^{-1}) were dissolved in distilled water and then sterilized by autoclaving:
59 NaCl (20), $Na_2S_2O_3 \cdot 5H_2O$ (5), $MgCl_2 \cdot 6H_2O$ (3), $MgSO_4 \cdot 7H_2O$ (0.3), $CaCl_2 \cdot 2H_2O$
60 (0.1), NH_4Cl (0.1), KH_2PO_4 (0.1) and KCl (0.1). To the autoclaved and cooled salt
61 solution, the following stock solutions (ml l^{-1}) were aseptically added: trace element
62 solution (1), selenite-tungstate solution (1), vitamin mixture solution (1) and 1M $NaHCO_3$
63 solution (30). The vitamin mixture solution consisted of the followings (mg l^{-1}): biotin
64 (20), folic acid (20), pyridoxine-HCl (100), thiamine-HCl·$2H_2O$ (50), riboflavin (50),

65 nicotinic acid (50), calcium D(+) pantothenate (50), 4-Aminobenzoic acid (50), lipoic
66 acid (50) and cyanocobalamine (1). The other stock solutions were prepared as described
67 previously (Widdel & Bak, 1992). Finally, pH of the medium was adjusted to 7.0–7.2
68 with HCl. From the enrichment culture established, pure culture of strain H1576^T was
69 obtained by repeated serial dilution with the basal medium. The enrichment and isolation
70 were performed at 15°C in the dark.

71 Phenotypic characteristics of strain H1576^T were investigate by culturing the
72 strain at 15°C in the basal medium, unless otherwise specified. Cell morphology was
73 observed with phase-contrast light microscopy, and Gram-stain test was conducted with
74 a kit (Fluka). Cellular fatty acid profile was obtained with the Sherlock Microbial
75 Identification System (MIDI) version 6.0 (database; TSBA6).

76 To determine upper and lower limits of temperature for growth, strain H1576^T
77 was inoculated into the basal medium and incubated at 0, 3, 5, 8, 13, 15, 18, 22, 25, 28,
78 30 and 32°C. Effect of NaCl concentration on growth was examined by using media
79 modified from the basal medium, with lowered concentration of $MgCl_2 \cdot 6H_2O$ (0.2 g l^{-1})
80 and varying concentrations of NaCl (0.0, 0.5, 1.0, 2.0, 3.0, 4.0, 5.0 % and 6.0 w/v).
81 Effect of pH on growth was tested with media of various pH which were prepared as
82 below. The media commonly contained the following constituents (l^{-1}): 20 g NaCl, 5 g

83 $Na_2S_2O_3 \cdot 5H_2O$, 1 g $NaHCO_3$, 0.2 g $MgCl_2 \cdot 6H_2O$, 0.1 g $CaCl_2 \cdot 2H_2O$, 0.1 g NH_4Cl, 0.1 g
84 KH_2PO_4, 0.1 g KCl, 1 ml trace element solution, 1 ml selenite-tungstate solution and 1 ml
85 vitamin mixture solution. Each medium of varying pH contained one of buffering
86 reagents listed below (at a final concentration of 20 mM), along with NaOH for pH
87 adjustment. Tested pH and buffering reagents were as follows; pH 5.8, 5.9, 6.0, 6.1, 6.2,
88 6.3, 6.4, 6.5, 6.7, 6.8, 6.9, 7.1, 7.2, 7.4 and 7.7 with MES; pH 6.6, 6.9 and 7.2 with PIPES;
89 pH 7.0, 7.2, 7.3 and 7.6 with MOPS. All ingredients were mixed and then sterilized by
90 filtration.

91 Utilization of electron donors was tested with the basal medium, by replacing
92 thiosulfate with one of the followings (mM); sulfide (2), pyruvate (5), lactate (5), acetate
93 (5), propionate (2.5), succinate (2.5), fumarate (2.5), malate (2.5), butyrate (2.5), benzoate
94 (2.5), isobutyrate (2.5), methanol (5), ethanol (2.5), formate (5), citrate (5), glucose (2.5),
95 xylose (2.5), phenol (2), *m*-cresol (1). As insoluble substrates, elemental sulfur (0.5 g l^{-1})
96 and hydrogen gas (air/H_2; 2 : 1, v/v; 150 kPa total pressure) were also tested with the
97 thiosulfate-free basal medium. Utilization of electron acceptors was tested with the basal
98 medium supplemented with nitrite (2 mM) or nitrate (5, 10 mM), under atmosphere of N_2
99 and CO_2 (80% and 20% in volume, respectively).

100 The novel isolate was subjected to whole genome sequencing, with the PacBio

RS II platform. From linear contigs obtained, circular chromosome and plasmid were manually reconstructed based on sequence alignment. The resulting complete genome sequence was subjected to comparative analysis with the closest relatives, by the TYGS web server (https://tygs.dsmz.de). In the TYGS, the Type (Strain) Genome Server, relatives of the subjected genome were automatically identified for subsequent genome-based phylogenetic analysis and calculation of digital DNA-DNA hybridization (dDDH) values (Meier-Kolthoff & Göker, 2019). Phylogenetic analysis was also conducted with the 16S rRNA gene identified in the genome, by using MEGA version 11 (Tamura et al., 2021). The reference sequences of *Sulfurimonas* species were retrieved from LPSN (accessed on 06 July 2022). The sequences of strain H1576^{T} and references were aligned with the MUSCLE algorithm. As an outgroup, *Sulfuricurvum kujiense* YK-1^{T} was included in the alignment. The best substitution model with the lowest Bayesian Information Criterion score was selected by the model selection tool in MEGA. Phylogenic tree was constructed with the selected model by excluding positions with gaps. Values of average nucleotide identity (ANI) between strain H1576^{T} and type strains of *Sulfurimonas* species were computed by ANI calculator available in the EzBioCloud, based on the OrthoANIu algorithm (Yoon et al., 2017).

Results

Cells of the novel isolate, strain H1576^T, were Gram-stain-negative, motile, rod-shaped, 1.5–2.7 μm in width, 0.6–0.7 μm in length. The strain grew at 3–22°C with optimum growth at 13–15°C. At 15°C, growth was observed at pH range of 6.0–7.4, with optimum growth at pH of 6.6–6.8. Growth was observed in the presence of 2–5% (w/v) NaCl. The cellular fatty acid profile of strain H1576^T is shown in Table S1. In the profile, summed feature 3 ($C_{16:1}\omega7c$ and/or $C_{16:1}\omega6c$) and $C_{16:0}$ were predominant, accounting for 65.5% and 21.9%, respectively.

Chemolithoautotrophic growth of strain H1576^T was supported by thiosulfate, sulfide and elemental sulfur, but not by H_2 gas. None of the tested organic substrate supported aerobic growth of the strain. As sole electron acceptor for thiosulfate oxidation, nitrate and nitrite did not support anaerobic growth of strain H1576^T.

The reconstructed genome of strain H1576^T consists of a circular chromosome and a plasmid, with length of 2.76 Mbp and 81.9 kbp, respectively. The G+C contents of the chromosome and plasmid are 34.8% and 32.8%, respectively. By analyzing the genome with the TYGS platform, it was revealed that the closest relatives of strain H1576^T are *Sulfurimonas* species. Genome-based phylogenetic analysis by the TYGS indicated that strain H1576^T belongs to the genus *Sulfurimonas*, but not to any known

137 species (Fig. S1). The calculated values of dDDH and ANI indicated strain H1576^T should
138 not be affiliated to any *Sulfurimonas* species previously proposed (Table 1). Phylogenetic
139 analysis was also conducted with the 16S rRNA gene identified in the genome. The
140 generated phylogenetic tree indicated that strain H1576^T is phylogenetically distinct from
141 all type strains of the genus (Fig. 1). The genome of H1576^T has been incorporated in the
142 genome taxonomy database (GTDB), which provides genome-based taxonomy
143 framework on the basis of conserved proteins (Parks et al., 2018). In the latest release of
144 the GTDB (07-RS207), strain H1576^T is classified into a *Sulfurimonas* species which
145 encompasses no other organisms. All these analyses consistently indicate that strain
146 H1576^T is representative of a new species in the genus *Sulfurimonas*.
147
148 **Conclusion**
149 The genomic analyses of different approaches consistently indicated that strain
150 H1576^T should be classified into a new species of the genus *Sulfurimonas*. Within the
151 genus, strain H1576^T is differentiated from the type strains of the other species by a
152 unique combination of phenotypic characteristics (Table 1). On the basis of these results,
153 H1576^T is proposed to be assigned to a new species, with the name *Sulfurimonas aquatica*
154 sp. nov.

155

156 **Description of *Sulfurimonas aquatica* sp. nov.**

157 ***Sulfurimonas aquatica*** (a.qua'ti.ca. L. fem. adj. *aquatica*, aquatic).

158 Cells and rod-shaped, motile, 1.5–2.7 μm in length and 0.6–0.7 μm in width. Gram-stain
159 -negative. Grows chemolithoautotrophically by oxidizing thiosulfate, sulfide and
160 elemental sulfur. Hydrogen gas is not used as electron donor. Aerobic. Nitrate and nitrite
161 do not support anaerobic growth when thiosulfate is provided as the sole electron donor.
162 Grows at 3–22°C with an optimum growth at 13–15°C. The pH range for growth is 6.0–
163 7.4, with an optimum pH range of 6.6–6.8. Grows with 2–5% NaCl (optimum 2–3%).
164 Predominant fatty acid is $C_{16:1}$. G + C content of genomic DNA of the type strain is 34.7
165 mol%.

166 The type strain H1576^T (= BCRC 81254^T = JCM 35004^T) was isolated from
167 water of a brackish lake in Japan.

168 The GenBank/EMBL/DDBJ accession numbers for the chromosome and
169 plasmid of type strain are CP046072 and CP046073, respectively.

170

References

Cai L, Shao M-F, Zhang T (2014) Non-contiguous finished genome sequence and description of *Sulfurimonas hongkongensis* sp. nov., a strictly anaerobic denitrifying, hydrogen- and sulfur-oxidizing chemolithoautotroph isolated from marine sediment. Stand Genomic Sci 9:1302–1310. https://doi.org/10.4056/sigs.4948668

Fu C, Li J, Lv X, Song W, Zhang X (2020) Operation performance and microbial community of sulfur-based autotrophic denitrification sludge with different sulfur sources. Environ Geochem Health 42:1009–1020. https://doi.org/10.1007/s10653-019-00482-5

Han Y, Perner M (2015) The globally widespread genus *Sulfurimonas*: versatile energy metabolisms and adaptations to redox clines. Front Microbiol 6:989. https://doi.org/10.3389/fmicb.2015.00989

Haosagul S, Oaew S, Prommeenate P, Sawasdee V, Boonyawanich S, Pisutpaisal N (2021) Profile of sulfur oxidizing bacteria in full-scale biotrickling filter to remove H_2S in biogas from in cassava starch industry. Energy Rep 7:677–685. https://doi.org/10.1016/j.egyr.2021.07.086

Henkel JV, Vogts A, Werner J, Neu TR, Spröer C, Bunk B, Schulz-Vogt HN (2021) *Candidatus* Sulfurimonas marisnigri sp. nov. and *Candidatus* Sulfurimonas baltica sp. nov., thiotrophic manganese oxide reducing chemolithoautotrophs of the class

190 *Campylobacteria* isolated from the pelagic redoxclines of the Black Sea and the Baltic
191 Sea. Syst Appl Microbiol 44:126155. https://doi.org/10.1016/j.syapm.2020.126155
192 Hu Q, Wang S, Lai Q, Shao Z, Jiang L (2021) *Sulfurimonas indica* sp. nov., a hydrogen-
193 and sulfur-oxidizing chemolithoautotroph isolated from a hydrothermal sulfide
194 chimney in the Northwest Indian Ocean. Int J Syst Evol Microb 71:004575.
195 https://doi.org/10.1099/ijsem.0.004575
196 Inagaki F, Takai K, Kobayashi H, Nealson KH, Horikoshi K (2003) *Sulfurimonas*
197 *autotrophica* gen. nov., sp. nov., a novel sulfur-oxidizing ε-proteobacterium isolated
198 from hydrothermal sediments in the Mid-Okinawa Trough. Int J Syst Evo Microbiol
199 53:1801–1805. https://doi: 10.1099/ijs.0.02682-0
200 Kubo K, Kojima H, Fukui M (2014) Vertical distribution of major sulfate-reducing
201 bacteria in a shallow eutrophic meromictic lake. Syst Appl Microbiol 37:510–519.
202 https://doi.org/10.1016/j.syapm.2014.05.008
203 Labrenz M, Grote J, Mammitzsch K, Boschker HT, Laue M, Jost G et al. (2013)
204 *Sulfurimonas gotlandic*a sp. nov., a chemoautotrophic and psychrotolerant
205 epsilonproteobacterium isolated from a pelagic redoxcline, and an emended
206 description of the genus *Sulfurimonas*. Int J Syst Evol Microbiol 63:4141–4148.
207 https://doi: 10.1099/ijs.0.048827-0

208 Meier-Kolthoff JP, Göker M (2019) TYGS is an automated high-throughput platform for
209 state-of-the-art genome-based taxonomy. Nat Commun 10:2182.
210 https://doi.org/10.1038/s41467-019-10210-3
211 Parks DH, Chuvochina M, Waite DW, Rinke C, Skarshewski A, Chaumeil PA,
212 Hugenholtz P (2018) A standardized bacterial taxonomy based on genome phylogeny
213 substantially revises the tree of life. Nat Biotechnol 36:996–1004.
214 https://doi.org/10.1038/nbt.4229
215 Parte AC, Sardà Carbasse J, Meier-Kolthoff JP, Reimer LC, Göker M (2020) List of
216 Prokaryotic names with Standing in Nomenclature (LPSN) moves to the DSMZ. Int
217 J Syst Evol Microbiol 70:5607–5612. https://doi.org/10.1099/ijsem.0.004332
218 Ratnikova NM, Slobodkin AI, Merkel AY, Kopitsyn DS, Kevbrin VV, Bonch-
219 Osmolovskaya EA, Slobodkina GB (2019) *Sulfurimonas crateris* sp. nov., a
220 facultative anaerobic sulfur-oxidizing chemolithoautotrophic bacterium isolated from
221 a terrestrial mud volcano. Int J Syst Evol Microbiol 70:487–492
222 https://doi.org/10.1099/ijsem.0.003779
223 Takai K, Suzuki MT, Nakagawa S, Miyazaki M, Suzuki Y, Inagaki F et al. (2006)
224 *Sulfurimonas paralvinellae* sp. nov., a novel mesophilic, hydrogen- and sulfur-
225 oxidizing chemolithoautotroph within the *Epsilonproteobacteria* isolated from a

226 deep-sea hydrothermal vent polychaete nest, reclassification of *Thiomicrospira*
227 *denitrificans* as *Sulfurimonas denitrificans* comb. nov. and emended description of
228 the genus *Sulfurimonas*. Int J Syst Evol Microbiol 56:1725–1733.
229 https://doi.org/10.1099/ijs.0.64255-0
230 Tamura K, Stecher G, Kumar S (2021) MEGA11: Molecular Evolutionary Genetics
231 Analysis version 11. Mol Biol Evol 38:3022–3027.
232 https://doi.org/10.1093/molbev/msab120
233 Timmer-Ten Hoor A (1975) A new type of thiosulphate oxidizing, nitrate reducing
234 microorganism: *Thiomicrospira denitrificans* sp. nov. Netherlands J Sea Res 9:344–
235 350. http://dx.doi.org/10.1016/0077-7579(75)90008-3
236 Wang S, Jiang L, Liu X, Yang S, Shao Z (2020) *Sulfurimonas xiamenensis* sp. nov. and
237 *Sulfurimonas lithotrophica* sp. nov., hydrogen and sulfur-oxidizing
238 chemolithoautotrophs within the *Epsilonproteobacteria* isolated from coastal
239 sediments, and an emended description of the genus. Int J Syst Evol Microbiol
240 70:2657–2663. https://doi.org/10.1099/ijsem.0.004087
241 Wang S, Jiang L, Hu Q, Cui L, Zhu B, Fu X, Lai Q, Shao Z, Yang, S (2021a)
242 Characterization of *Sulfurimonas hydrogeniphila* sp. nov., a novel bacterium
243 predominant in deep-sea hydrothermal vents and comparative genomic analyses of

the genus *Sulfurimonas*. Front Microbiol 12:626705. https://doi.org/10.3389/fmicb.2021.626705

Wang S, Shao Z, Lai Q, Liu X, Xie S, Jiang L, Yang S (2021) *Sulfurimonas sediminis* sp. nov., a novel hydrogen-and sulfur-oxidizing chemolithoautotroph isolated from a hydrothermal vent at the Longqi system, southwestern Indian ocean. Antonie van Leeuwenhoek 114:813–822. https://doi.org/10.1007/s10482-021-01560-4

Watanabe T, Kubo K, Kamei Y, Kojima H, Fukui M (2022) Dissimilatory microbial sulfur and methane metabolism in the water column of a shallow meromictic lake. Syst Appl Microbiol 45:126320. https://doi.org/10.1016/j.syapm.2022.126320

Widdel F, Bak F (1992) Gram-Negative Mesophilic Sulfate-Reducing Bacteria. In: The Prokaryotes. 2nd ed. vol. 4 New York: Springer-Verlag, pp 3352–3378

Wu J, Jiang X, Jin Z, Yang S, Zhang J (2020) The performance and microbial community in a slightly alkaline biotrickling filter for the removal of high concentration H_2S from biogas. Chemosphere 249:126127. https://doi.org/10.1016/j.chemosphere.2020.126127

Yoon S-H, Ha S-M, Lim J, Kwon S, 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

262

263 **Statements and Declarations**

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268

269 **Figure legends**

270 Fig. 1. Phylogenetic position of strain H1576^T within the genus *Sulfurimonas*, based on
271 the 16S rRNA gene sequences. This maximum likelihood tree was constructed based on
272 the Kimura 2-parameter model. All positions containing gaps and missing data were
273 eliminated, leaving 1099 positions in the final dataset. A discrete gamma distribution was
274 used to model differences in evolutionary rates among sites (5 categories, parameter =
275 0.3206). The rate variation model allowed for some sites to be invariable (68.83% sites).
276 Bar, substitutions per site. Numbers on nodes represent percentage values of 1000
277 bootstrap resampling.

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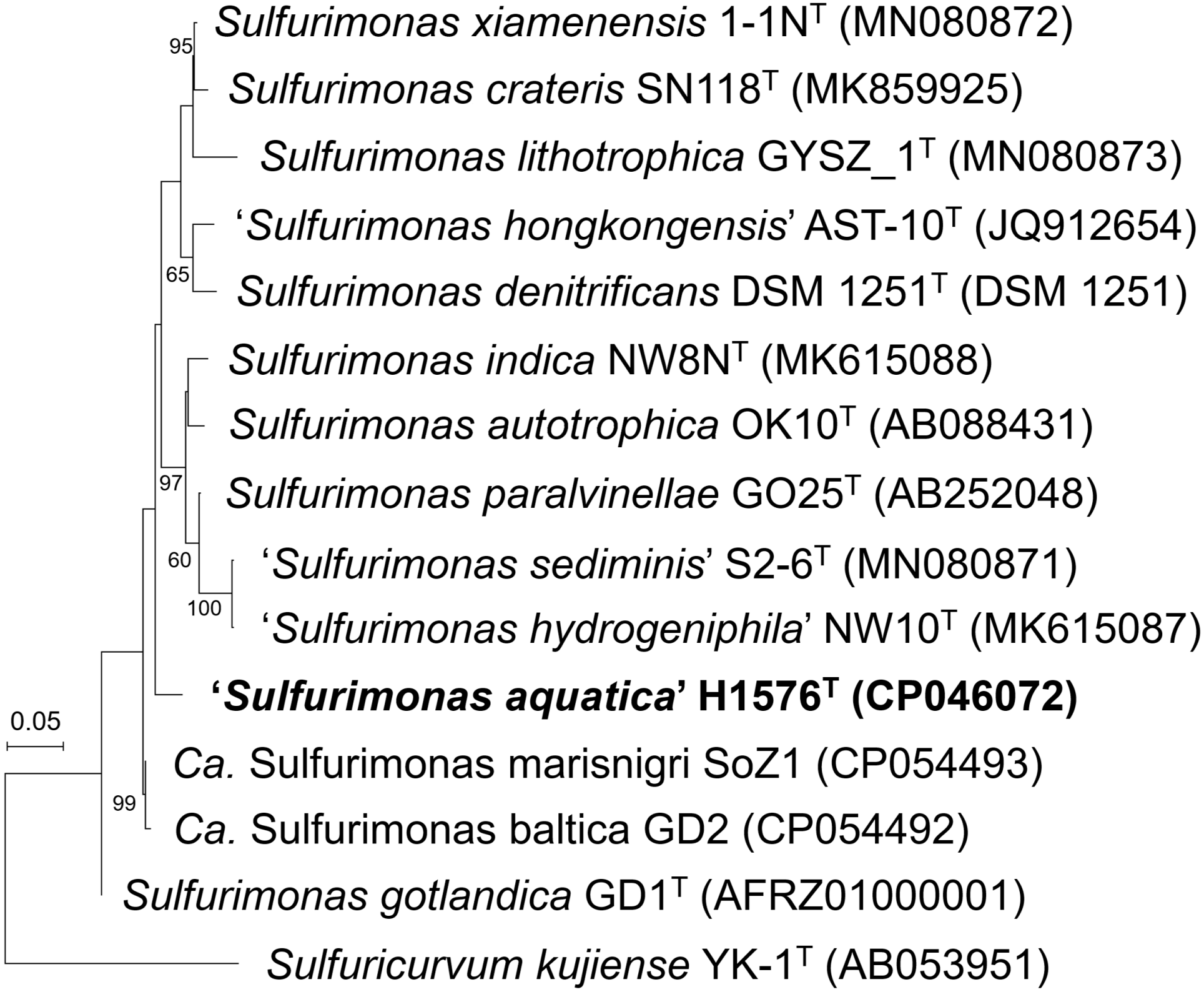
Sulfurimonas xiamenensis 1-1N^{T} (MN080872)
Sulfurimonas crateris SN118^{T} (MK859925)
Sulfurimonas lithotrophica GYSZ_1^{T} (MN080873)
'*Sulfurimonas hongkongensis*' AST-10^{T} (JQ912654)
Sulfurimonas denitrificans DSM 1251^{T} (DSM 1251)
Sulfurimonas indica NW8N^{T} (MK615088)
Sulfurimonas autotrophica OK10^{T} (AB088431)
Sulfurimonas paralvinellae GO25^{T} (AB252048)
'*Sulfurimonas sediminis*' S2-6^{T} (MN080871)
'*Sulfurimonas hydrogeniphila*' NW10^{T} (MK615087)
'*Sulfurimonas aquatica*' H1576^{T} (CP046072)
Ca. Sulfurimonas marisnigri SoZ1 (CP054493)
Ca. Sulfurimonas baltica GD2 (CP054492)
Sulfurimonas gotlandica GD1^{T} (AFRZ01000001)
Sulfuricurvum kujiense YK-1^{T} (AB053951)
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0.05

Table 1. Differential properties of strain H1576^T and strains representing *Sulfurimonas* species. Strains: 1, H1576^T; 2, *S. autotrophica* OK10^T (Inagaki et al., 2003); 3, *S. paralvinellae* GO25^T (Takai et al., 2006); 4, *S. denitrificans* DSM 1251^T (Timmer-Ten Hoor, 1975); 5, *S. gotlandica* GD1^T (Labrenz et al., 2013); 6, *S. crateris* SN118^T (Ratnikova et al., 2020); 7, *S. xiamenensis* 1-1N^T (Wang et al., 2020); 8, *S. lithotrophica* GYSG_1^T (Wang et al., 2020); 9, *S. indica* NW8N^T (Hu et al., 2021); 10, '*S. hongkongensis*' AST-10^T (Cai et al., 2014); 11, '*S. hydrogeniphila*' NW10^T (Wang et al., 2021a); 12, '*S. sediminis*' S2-6^T (Wang et al., 2021b); 13, *Ca*. S. marisnigri SoZ1 (Henkel et al., 2021); 14, *Ca*. S. baltica GD2 (Henkel et al., 2021). NR, not reported.

Strain:	1	2	3	4	5	6	7	8	9	10	11	12	13	14
Optimum temperature for growth (°C)	13-15	23-26	30	22	15	30	30	33	33	30	33	33	20	15
Growth at 5°C or lower	+	-	+	-	+	+	-	+	+	-	+	-	+	+
Growth at 35°C or higher	-	+	+	-	-	+	+	+	+	+	+	+	-	-
Optimum pH for growth	6.6-6.8	6.5	6.1	7.0	6.7-8.0	8.0	7.0	6.5	5.5	7.0-7.5	6.0-6.5	7.0	7.5-8.0	7.0-7.5
Growth at 8.5 or higher pH	-	+	+	NR	-	+	-	+	-	-	-	-	-	-
Growth at 5.5 or lower pH	-	+	+	NR	-	-	+	+	+	-	-	+	-	-
Growth by H_2 oxidation	-	-	+	-	+	-	+	+	+	+	+	+	+	+
Nitrate respiration	-	-	+	+	+	+	+	+	-	+	+	+	+	+
Nitrite respiration	-	-	-	+	+	+	-	-	-	-	-	-	-	-
dDDH with strain H1576^T (%)	100	19.4	18.5	19.1	20.1	18.2	21.1	18.6	18.9	19.0	18.7	19.1	20.2	20.4
ANI with strain H1576^T (%)	100	73.5	72.2	71.9	73.5	71.3	72.2	71.7	72.9	72.4	72.2	72.5	73.3	72.7