Stakelama pacifica gen. nov., sp. nov., a new member of the family *Sphingomonadaceae* isolated from the Pacific Ocean

Chuang Chen,¹ Qiang Zheng,¹ Ya-Nan Wang,^{1,2} Xiao-Jun Yan,³ Li-Kai Hao,¹ Xun Du² and Nianzhi Jiao¹

Correspondence

Nianzhi Jiao

Jiao@xmu.edu.cn

¹State Key Laboratory for Marine Environmental Science, Xiamen University, Xiamen 361005, PR China

²Key Laboratory of Microbial Engineering at the Institute of Biology, Henan Academy of Sciences, Zhengzhou 450008, PR China

³Key Laboratory of Applied Marine Biotechnology, Ministry of Education, Ningbo University, Ningbo 315211, PR China

A Gram reaction-negative, weakly motile, non-spore-forming, rod-shaped, aerobic bacterium designated strain JLT832^T was isolated from surface water of the central Pacific Ocean and formed yellow colonies on rich organic (RO) medium. The strain was oxidase-negative and catalase-positive. Acid was produced from mannitol, glucose, sucrose, lactose, sorbitol, maltose, (+)-trehalose and D-fructose. No acid was produced from D-(+)-xylose. The major cellular fatty acids of strain JLT832^T were C_{18:1}ω7c, C_{14:0} 2-OH and C_{16:0}. The major polar lipids were sphingoglycolipid, diphosphatidylglycerol, phosphatidylglycerol and phosphatidylethanolamine. Ubiquinone-10 and spermidine were present as the major quinone and polyamine, respectively. The genomic DNA G+C content of strain JLT832^T was 66.0 ± 0.5 mol%. Phylogenetic analysis based on 16S rRNA gene sequences indicated that the new isolate formed a tight branch within the family *Sphingomonadaceae* but was clearly separate from established genera in this family. The sequence similarities between the new isolate and type strains of established genera ranged from 90.5 to 94.9 %. Based on these data, strain JLT832^T constitutes a novel genus and species, for which the name *Stakelama pacifica* gen. nov., sp. nov. is proposed. The type strain of *Stakelama pacifica* is JLT832^T (=CGMCC 1.7294^T =LMG 24686^T).

The family *Sphingomonadaceae* was established by Kosako *et al.* (2000) and was further divided into families *Sphingomonadaceae* and *Erythrobacteraceae* by Lee *et al.* (2005). The genus *Sphingomonas* was first described by Yabuuchi *et al.* (1990) and was later divided into four genera, *Sphingomonas*, *Sphingobium*, *Novosphingobium* and *Sphingopyxis* (Takeuchi *et al.*, 2001), based on both phylogenetic analysis of 16S rRNA gene sequences and chemotaxonomic and phenotypic differences. Many novel genera of this clade have been described, such as *Sphingomonas*, *Sphingobium*, *Novosphingobium*, *Sphingopyxis*,

Zymomonas, *Sandaracinobacter*, *Sphingosinicella*, *Sandarakinorhabdus* and *Blastomonas*.

Strain JLT832^T was isolated from the surface of the Pacific Ocean at 174° 21.70' E 20° 34.44' N and formed a deep branch within the family *Sphingomonadaceae*. In this study, we characterized strain JLT832^T by using a polyphasic approach.

Strain JLT832^T was initially isolated with seawater medium (SW) containing (per litre seawater filtered by 0.2 µm film) 1 g peptone, 1 g yeast extract, 20 µg vitamin B₁₂ and 1 ml of a trace element solution (ES) as described by Drews (1983). The strain was identified by growing on modified rich organic (RO) medium (Yurkov *et al.*, 1999) containing (per litre distilled water) 1 g peptone, 1 g yeast extract, 20 g NaCl, 1 g sodium acetate, 0.3 g KCl, 0.5 g MgSO₄·7H₂O, 0.05 g CaCl₂·7H₂O, 0.3 g NH₄Cl, 0.3 g K₂HPO₄, 20 µg vitamin B₁₂ and 1.0 ml ES, at pH 8.0 and 28 °C.

Cell morphology was examined using light microscopy (BX61; Olympus) and TEM (H600; Hitachi). The Gram

Abbreviations: UPLC-Q-TOF-MS, ultra-performance liquid chromatography coupled with quadrupole time-of-flight mass spectrometry; MR, methyl red; VP, Voges–Proskauer.

The GenBank/EMBL/DDBJ accession number for the 16S rRNA gene sequence of strain JLT832^T is EU581829.

A transmission electron micrograph of negatively stained cells and polar lipid profiles of strain JLT832^T are available as supplementary material with the online version of this paper.

reaction was tested according to standard procedures (Gerhardt *et al.*, 1994). Catalase activity was determined by adding drops of 3% H_2O_2 to an overnight colony and assessed by the formation of bubbles. The pH and temperature ranges for growth were determined by adjusting the RO medium to a final pH of 5.5–9.5 (at intervals of 0.5 pH units) with HCl and NaOH, and by incubation at 0, 5, 10, 15, 25, 28, 30, 37 and 40 °C. Growth in NaCl 1–6% (w/v) (increments of 0.5%) was investigated in RO medium. Oxidase and urease activity, hydrolysis of casein, starch, gelatin and Tween 80 and reduction of nitrate and nitrite were determined as described by Dong & Cai (2001) on RO medium. Denitrification was tested by growing the strain anaerobically in the presence of nitrate (Zumft, 1992). H_2S production was tested as described by Bruns *et al.* (2001). Indole production, methyl red (MR) test and Voges–Proskauer (VP) reaction were determined as described by Denner *et al.* (2001).

The ability of strain JLT832^T to utilize substrates as sole carbon and energy sources was examined on RO medium without organic compounds at 28 °C for 7–14 days (Williams *et al.*, 1983). Acid production from carbohydrates was assessed using the procedures outlined by Cappuccino & Sherman (2002). Antibiotic susceptibility was tested by using the disc-diffusion plate (Kirby–Bauer) method according to Fraser & Jorgensen (1997) and Andrews (2008).

Cellular fatty acid composition was determined as described previously (Hu *et al.*, 2004) by growing cells of strain JLT832^T on marine agar 2216e (MA; Difco) for 2 days at 28 °C. Polyamines were extracted and derivatized as described by Busse & Auling (1988) and analysed by HPLC using the equipment previously described by Stolz *et al.* (2007). Polar lipids were identified by two-dimensional TLC and detection with sulfuric acid/ethanol (1:2, v/v) followed by heating at 150 °C for 3 min, according to the procedures described by Minnikin *et al.* (1984). Isoprenoid quinones were analysed as described by Hiraishi *et al.* (1998) using ultra-performance liquid chromatography coupled with quadrupole time-of-flight mass spectrometry (UPLC-Q-TOF-MS) in an electrospray ionization method (Romano *et al.*, 2006).

Genomic DNA was extracted following the method of Marmur (1961) from cells that were grown in RO medium for 2 days at 28 °C, washed and then resuspended in the buffer. Purity was assessed by measuring absorbance at A_{280}/A_{260} and A_{230}/A_{260} ratios (Johnson, 1994). The 16S rRNA gene was amplified and sequenced as described by Rainey *et al.* (1996) and sequences were aligned using the BLAST tool (<http://blast.ncbi.nlm.nih.gov/Blast.cgi>) (Altschul *et al.*, 1990) to determine the approximate phylogenetic affiliation. Phylogenetic trees were constructed using neighbour-joining and maximum-parsimony algorithms in MEGA 4.0

Fig. 1. Neighbour-joining phylogenetic tree based on 16S rRNA gene sequences showing the relationship between strain JLT832^T and representatives of the family Sphingomonadaceae. Bootstrap percentages (above 50%) from both neighbour-joining (above nodes) and maximum-parsimony (below nodes) are shown. Bar, 0.005 substitutions per nucleotide position.

software (Kumar *et al.*, 2004). DNA G+C contents were determined by thermal denaturation (Marmur & Doty, 1962) using DNA of *Escherichia coli* DH5 α as a standard for calibration of the T_m .

Strain JLT832^T cells were strictly aerobic, heterotrophic, Gram reaction-negative, short rods. Flagella were observed (Supplementary Fig. S1, available in IJSEM Online). Colonies were gold–yellow, glossy, opaque, circular and about 1 mm in diameter. JLT832^T grew in the presence of 0–5% (w/v) NaCl (optimum 2%), at 5–37 °C (optimum 28 °C) and pH 6–9 (optimum pH 8). Strain JLT832^T was positive for catalase, aerobic nitrate reduction, H₂S production and VP reaction and negative for oxidase, urease, aerobic nitrite reduction, anaerobic nitrate reduction, indole production, MR test and hydrolysis of gelatin, starch, Tween 80 and casein. Acid was produced from

mannitol, glucose, sucrose, lactose, sorbitol, maltose, (+)-trehalose and D-fructose. No acid was produced from (+)-D-xylose.

JLT832^T exhibited a polyamine pattern with the following compounds (per g dry weight): spermidine (17.6 μ mol), putrescine (0.2 μ mol), cadaverine (0.6 μ mol) and traces (<0.1 μ mol) of 1,3-diaminopropane and spermine. This polyamine pattern was in agreement with the traits reported for the genera *Blastomonas*, *Sphingobium*, *Novosphingobium* and *Sphingopyxis*, while the presence of homospermidine was restricted in species of *Zymomonas*, *Sphingosinicella* and *Sphingomonas* (Busse *et al.*, 1999; Takeuchi *et al.*, 2001; Hamana *et al.*, 2003). The major polar lipids were sphingoglycolipid, diphosphatidylglycerol, phosphatidylglycerol and phosphatidylethanolamine (Supplementary Fig. S2). Q-10 was the sole respiratory

Table 1. Differentiation of physiological and biochemical characteristics between strain JLT832^T and closely related species within the family *Sphingomonadaceae*

Taxa: 1, JLT832^T; 2, *Sphingosinicella microcystinivorans* JCM 13185^T; 3, *Sphingosinicella xenopeptidilytica* DSM 17130^T; 4, *Sphingosinicella soli* DSM 17328^T; 5, *Novosphingobium panipatense* CCM 7472^T; 6, *Novosphingobium mathurense* CCM 7473^T; 7, *Novosphingobium subarcticum* SMCC B0478^T; 8, *Zymomonas* species. Data in columns were taken from the following sources: 1–4, Maruyama *et al.* (2006), Geueke *et al.* (2007) and Yoon *et al.* (2008); 5–6, Gupta *et al.* (2009); 7, Nohynek *et al.* (1996) and Lim *et al.* (2007); 8, De Ley & Swings (1976) and Swings & De Ley (1977). All strains were negative for Gram stain, gelatin hydrolysis and indole production. Q-10 was the respiratory quinone for all the strains. w, Weak reaction; ND, no data.

Characteristic	1	2	3	4	5	6	7	8
Motility	+	+	ND	+	–	ND	ND	–/+
Oxidase	–	+	+	+	–	–	–	–
Catalase	+	+	+	+	–	–	+	+
Hydrolysis of:								
Starch	–	ND	–	–	–	–	ND	–
Tween 80	–	–	–	+	+	+	+	–
Aerobic nitrate reduction	+	–	+	–	w	–	+	–
H ₂ S production	+	–	+	–	ND	ND	–	–/+
Assimilation of:								
(+)-D-Xylose	+	–	–	ND	–	+	+	–
Maltose	+	–	–	ND	+	+	+	–
Sucrose	w	–	–	–	w	+	–	+
D-Fructose	–	–	–	–	+	+	+	+
Glucose	w	–	–	–	+	+	+	+
(+)-D-Galactose	+	–	–	–	+	+	+	–
Lactose	w	–	–	–	+	ND	ND	–
(+)-Trehalose	+	–	–	–	+	+	+	–
Sorbitol	+	–	–	–	+	–	–	–
Inositol	+	–	–	–	–	–	–	–
Glycerol	+	–	–	–	–	–	+/-	–
Mannitol	+	–	–	–	–	–	+	–
D-Gluconate	+	–	–	ND	ND	ND	–	+
Trisodium citrate	–	w	+	–	ND	ND	+	–
Acetic acid	+	–	–	–	+	ND	ND	ND
Malonic acid	+	+	+	–	–	w	–	ND
(–)-L-Malic acid	w	–	–	ND	ND	ND	+	–
L-Proline	w	+	+	–	+	+	+	–/+
DNA G+C content (mol%)	66.0 $\pm$ 0.5	63.6–63.7	65	65	ND	ND	66 $\pm$ 1.0	48.5 $\pm$ 1.0

quinone. Cellular fatty acids were $C_{18:1\omega7c}$ (72.88 %), $C_{14:0}$ 2-OH (11.93 %), $C_{16:0}$ (11.53 %), $C_{18:1\omega5c}$ (2.02 %) and $C_{16:1\omega7c}$ (1.64 %) (Supplementary Table S1).

The DNA G + C content of JLT832^T was 66 ± 0.5 mol%. The complete 16S rRNA gene sequence (1412 bp) of strain JLT832^T was determined. Phylogenetic trees based on 16S rRNA gene sequences (1374 bp compared) showed that strain JLT832^T forms a distinct phylogenetic lineage within the family *Sphingomonadaceae* (Fig. 1) and exhibits the highest sequence similarity to *Novosphingobium subarcticum* SMCC B0478^T (94.9 %), followed by *Novosphingobium mathurense* SM 117^T (94.3 %) and *Sphingosinicella microcystinivorans* JCM 13185^T (94.3 %).

No currently recognized species in the genera *Novosphingobium*, *Sphingosinicella* and *Zymomonas* utilize sorbitol except *Novosphingobium panipatense* and *N. subarcticum*. *N. subarcticum* only utilized mannitol, while strain JLT832^T could utilize sorbitol, mannitol, *myo*-inositol and glycerol.

A comparison of differential characteristics of strain JLT832^T and members of related genera *Novosphingobium*, *Sphingosinicella* and *Zymomonas* are listed in Table 1. The morphological, physiological and biochemical characteristics of strain JLT832^T indicate that the new isolate represents a novel genus and species of the family *Sphingomonadaceae*, for which the name *Stakelama pacifica* gen. nov., sp. nov. is proposed.

Description of *Stakelama* gen. nov.

Stakelama (Sta.ke.la'ma. N.L. fem. n. *Stakelama* arbitrary name derived from State Key Laboratory of Marine Environment Science).

Cells are Gram reaction-negative, weakly motile, non-spore-forming rods. Colonies are gold-yellow. Strictly aerobic. Positive for catalase and negative for oxidase. The major fatty acids are $C_{18:1\omega7c}$, $C_{14:0}$ 2-OH and $C_{16:0}$. The sole respiratory quinone is Q-10. The DNA G + C content of the type strain of the type species is 66 ± 0.5 mol%. The type species is *Stakelama pacifica*.

Description of *Stakelama pacifica* sp. nov.

Stakelama pacifica (pa.ci'fi.ca. L. fem. adj. *pacifica* peaceful, referring to the Pacific Ocean, the origin of the type strain).

The following properties are shown in addition to those given in the genus description. Cells are $0.5\text{--}0.8 \times 1.5\text{--}2.0$ μm , and have a flagellum. Colonies on RO medium are circular, slightly raised, smooth and 1 mm in diameter after incubation for 24 h at 28 °C. Growth occurs at 5–37 °C (optimum 28 °C), pH 6–9 (optimum pH 8) and in the presence of 0–5 % (w/v) NaCl (optimum 2 %). Positive for catalase, aerobic nitrate reduction, H_2S production and the VP reaction. Negative for oxidase, urease, aerobic nitrite reduction, anaerobic nitrate reduction, indole production, the MR test and hydrolysis of gelatin, starch, Tween 80 and casein. (+)-D-Xylose, maltose, sorbitol,

lactose, (+)-trehalose, cellobiose, lactic acid, sodium D-gluconate, inositol, glycerol, mannitol, acetic acid, ethanol and malonic acid are utilized. Sucrose, glucose, (+)-D-galactose, (–)-L-malic acid, L-proline and L-threonine are used weakly. (–)-L-Sorbose, D-fructose, dextrin, trisodium citrate, oxalic acid, L-alanine, L-lysine, glycine and L-arginine are not utilized. The type strain was sensitive (μg per disc) to ampicillin (10), penicillin (10), tetracycline (30), chloramphenicol (30), erythromycin (15), cefoperazone (75), cefotaxime (30), ciprofloxacin (5), clarithromycin (15), gentamicin (120) and streptomycin (300); resistant to clindamycin (2), sulfamethoxazole (300), vancomycin (30) and cefalothin (30).

The type strain, JLT832^T (=CGMCC 1.7294^T =LMG 24686^T), was isolated from surface water at 174° 21.70' E 20° 34.44' N in the Pacific Ocean.

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References

- Altschul, S. F., Gish, W., Miller, W., Myers, E. W. & Lipman, D. J. (1990). Basic local alignment search tool. *J Mol Biol* **215**, 403–410.
- Andrews, J. M. (2008). BSAC standardized disc susceptibility testing method (Version 7). *J Antimicrob Chemother* **62**, 256–278.
- Bruns, A., Rohde, M. & Berthe-Corti, L. (2001). *Muricauda ruestringensis* gen. nov., sp. nov., a facultatively anaerobic, appendaged bacterium from German North Sea intertidal sediment. *Int J Syst Evol Microbiol* **51**, 1997–2006.
- Busse, H.-J. & Auling, G. (1988). Polyamine pattern as a chemotaxonomic marker within the *Proteobacteria*. *Syst Appl Microbiol* **11**, 1–8.
- Busse, H. J., Kämpfer, P. & Denner, E. B. (1999). Chemotaxonomic characterisation of *Sphingomonas*. *J Ind Microbiol Biotechnol* **23**, 242–251.
- Cappuccino, J. G. & Sherman, N. (2002). *Microbiology: a Laboratory Manual*, 6th edn. San Francisco: Pearson Education, Inc. and Benjamin Cummings.
- De Ley, J. & Swings, J. (1976). Phenotypic description, numerical analysis and a proposal of an improved taxonomy and nomenclature of the genus *Zymomonas* Kluyver and van Niel 1936. *Int J Syst Bacteriol* **26**, 146–157.
- Denner, E. B., Paukner, S., Kämpfer, P., Moore, E. R., Abraham, W.-R., Busse, H.-J., Wanner, G. & Lubitz, W. (2001). Description of *Sphingomonas pituitosa* sp. nov., a novel polysaccharide-producing sphingomonad. *Int J Syst Evol Microbiol* **51**, 827–841.
- Dong, X.-Z. & Cai, M.-Y. (2001). *Determinative Manual for Routine Bacteriology*. Beijing: Scientific Press (in Chinese).
- Drews, G. (1983). *Mikrobiologisches Praktikum*. Berlin, Germany: Springer (in German).
- Fraser, S. L. & Jorgensen, J. H. (1997). Reappraisal of the antimicrobial susceptibilities of *Chryseobacterium* and *Flavobacterium* species and

methods for reliable susceptibility testing. *Antimicrob Agents Chemother* **41**, 2738–2741.

Gerhardt, P., Murray, R. G. E., Wood, W. A. & Krieg, N. R. (editors) (1994). *Methods for General and Molecular Bacteriology*. Washington, DC: American Society for Microbiology.

Geueke, B., Busse, H.-J., Fleishmann, T., Kämpfer, P. & Kohler, H.-P. E. (2007). Description of *Sphingosinicella xenopeptidilytica* sp. nov., a β -peptide-degrading species, and emended descriptions of the genus *Sphingosinicella* and *Sphingosinicella microcystinivorans*. *Int J Syst Evol Microbiol* **57**, 107–113.

Gupta, S. K., Lal, D. & Lal, R. (2009). *Novosphingobium panipatense* sp. nov. and *Novosphingobium matheurense* sp. nov., from oil-contaminated soil. *Int J Syst Evol Microbiol* **59**, 156–161.

Hamana, K., Sakamoto, A., Tachiyanagi, S., Terauchi, E. & Takeuchi, M. (2003). Polyamine profiles of some members of the alpha subclass of the *Proteobacteria*: polyamine analysis of 20 recently described genera. *Microbiol Cult Coll* **19**, 13–21.

Hiraishi, A., Ueda, Y. & Ishihara, J. (1998). Quinone profiling of bacterial communities in natural and synthetic sewage activated sludge for enhanced phosphate removal. *Appl Environ Microbiol* **64**, 992–998.

Hu, Y.-T., Zhou, P.-J., Zhou, Y.-G., Liu, Z.-H. & Liu, S.-J. (2004). *Xaccharothrix xingjiangensis* sp. nov., a pyrene-degrading actinomycete isolated from Tianchi Lake, Xinjiang, China. *Int J Syst Evol Microbiol* **54**, 2091–2094.

Johnson, J. L. (1994). Similarity analysis of DNAs. In *Methods for General and Molecular Bacteriology*, pp. 655–681. Edited by P. E. Gerhardt, R. G. Murray, W. A. Wood & N. R. Krieg. Washington, DC: American Society for Microbiology.

Kosako, Y., Yabuuchi, E., Naka, T., Fujiwara, N. & Kobayashi, K. (2000). Proposal of *Sphingomonadaceae* fam. nov., consisting of *Sphingomonas* Yabuuchi et al. 1990, *Erythrobacter* Shiba and Shimidu 1982, *Erythromicrobium* Yurkove et al. 1994, *Porphyrobacter* Fuerst et al. 1993, *Zymomonas* Kluyver and van Niel 1936, and *Sandaracinobacter* Yurkove et al. 1997, with the type genus *Sphingomonas* Yabuuchi et al. 1990. *Microbiol Immunol* **44**, 563–575.

Kumar, S., Tamura, K. & Nei, M. (2004). MEGA3: Integrated software for molecular evolutionary genetics analysis and sequence alignment. *Brief Bioinform* **5**, 150–163.

Lee, K.-B., Liu, C.-T., Anzai, Y., Kim, H., Aono, T. & Oyaizu, H. (2005). The hierarchical system of the 'Alphaproteobacteria': description of *Hyphomonadaceae* fam. nov., *Xanthobacteraceae* fam. nov. and *Erythrobacteraceae* fam. nov. *Int J Syst Evol Microbiol* **55**, 1907–1919.

Lim, Y. W., Moon, E. Y. & Chun, J. (2007). Reclassification of *Flavobacterium resinovorum* Delaporte and Daste 1956 as *Novosphingobium resinovorum* comb. nov., with *Novosphingobium subarcticum* (Nohynek et al. 1996) Takeuchi et al. 2001 as a later heterotypic synonym. *Int J Syst Evol Microbiol* **57**, 1906–1908.

Marmur, J. (1961). A procedure for the isolation of deoxyribonucleic acid from microorganisms. *J Mol Biol* **3**, 208–218.

Marmur, J. & Doty, P. (1962). Determination of the base composition of deoxyribonucleic acid from thermal denaturation temperature. *J Mol Biol* **5**, 109–118.

Maruyama, T., Park, H.-D., Ozawa, K., Tanaka, Y., Sumino, T., Hamana, K., Hiraishi, A. & Kato, K. (2006). *Sphingosinicella microcystinivorans* gen. nov., sp. nov., a microcystin-degrading bacterium. *Int J Syst Evol Microbiol* **56**, 85–89.

Minnikin, D. E., O'Donnell, A. G., Goodfellow, M., Alderson, G., Athalye, M., Schaal, A. & Parlett, J. H. (1984). An integrated procedure for the extraction of bacterial isoprenoid quinones and polar lipids. *J Microbiol Methods* **2**, 233–241.

Nohynek, L. J., Nurmiaho-Lassila, E.-L., Suhonen, E. L., Busse, H.-J., Mohammadi, M., Hantula, J., Rainey, F. & Salkinoja-Salonen, M. S. (1996). Description of chlorophenol-degrading *Pseudomonas* sp. strains KF1^T, KF3 and NK1 as a new species of the genus *Sphingomonas*, *Sphingomonas subarctica* sp. nov. *Int J Syst Bacteriol* **46**, 1042–1055.

Rainey, F. A., Ward-Rainey, N., Kroppenstedt, R. M. & Stackebrandt, E. (1996). The genus *Nocardiopsis* represents a phylogenetically coherent taxon and a distinct actinomycete lineage: proposal of *Nocardiopsaceae* fam. nov. *Int J Syst Bacteriol* **46**, 1088–1092.

Romano, I., Lama, L., Nicolaus, B., Poli, A., Gambacorta, A. & Giordano, A. (2006). *Halomonas alkaliphila* sp. nov., a novel halotolerant alkaliphilic bacterium isolated from a salt pool in Campania (Italy). *J Gen Appl Microbiol* **52**, 339–348.

Stolz, A., Busse, H.-J. & Kämpfer, P. (2007). *Pseudomonas knackmussii* sp. nov. *Int J Syst Evol Microbiol* **57**, 572–576.

Swings, J. & De Ley, J. (1977). The biology of *Zymomonas*. *Bacteriol Rev* **41**, 1–46.

Takeuchi, M., Hamana, K. & Hiraishi, A. (2001). Proposal of the genus *Sphingomonas sensu stricto* and three new genera, *Sphingobium*, *Novosphingobium* and *Sphingopyxis*, on the basis of phylogenetic and chemotaxonomic analyses. *Int J Syst Evol Microbiol* **51**, 1405–1417.

Williams, S. T., Goodfellow, M., Alderson, G., Wellington, E. M. H., Sneath, P. H. A. & Sackin, M. J. (1983). Numerical classification of *Streptomyces* and related genera. *J Gen Microbiol* **129**, 1743–1813.

Yabuuchi, E., Yano, I., Oyaizu, H., Hashimoto, Y., Ezaki, T. & Yamamoto, H. (1990). Proposals of *Sphingomonas paucimobilis* gen. nov. and comb. nov., *Sphingomonas parapaucimobilis* sp. nov., *Sphingomonas yanoikuyae* sp. nov., *Sphingomonas adhaesiva* sp. nov., *Sphingomonas capsulata* comb. nov., and two genospecies of the genus *Sphingomonas*. *Microbiol Immunol* **34**, 99–119.

Yoon, J.-H., Kang, S.-J., Lee, J.-S., Nam, S.-W., Kim, W. & Oh, T.-K. (2008). *Sphingosinicella soli* sp. nov., isolated from an alkaline soil in Korea. *Int J Syst Evol Microbiol* **58**, 173–177.

Yurkov, V. V., Krieger, S., Stackebrandt, E. & Beatty, J. T. (1999). *Cirtomicrobium bathyomarinum*, a novel aerobic bacterium isolated from deep-sea hydrothermal vent plume waters that contains photosynthetic pigment-protein complexes. *J Bacteriol* **181**, 4517–4525.

Zumft, W. G. (1992). The denitrifying bacteria. In *The Prokaryotes*, 2nd edn. pp. 554–582. Edited by A. Balows, H. G. Trüper, M. Dworkin, W. Harder & K. H. Schleifer. New York: Springer Verlag.