Bowmanella denitrificans gen. nov., sp. nov., a denitrifying bacterium isolated from seawater from An-Ping Harbour, Taiwan

Wen Dar Jean,[1] Jwo-Sheng Chen,[2] Yu-Te Lin[3] and Wung Yang Shieh[3]

Correspondence
Wung Yang Shieh
winyang@ntu.edu.tw

[1]Center for General Education, Leader University, No. 188, Sec. 5, An-Chung Road, Tainan, Taiwan

[2]School of Medicine, China Medical University, No. 91, Shyue-Shyh Road, Taichung, Taiwan

[3]Institute of Oceanography, National Taiwan University, PO Box 23-13, Taipei, Taiwan

A heterotrophic, non-fermentative, denitrifying isolate, designated strain BD1^T, was obtained from a seawater sample collected in the shallow coastal region of An-Ping Harbour, Tainan, Taiwan. The cells of strain BD1^T were Gram-negative. Cells grown in broth cultures were curved rods that were motile by means of a single polar flagellum. Growth occurred between 10 and 40 °C, with an optimum at 30–35 °C. Strain BD1^T grew in NaCl levels of 0–10 %, with better growth occurring at 1–3 %. It grew aerobically and could achieve anaerobic growth by adopting a denitrifying metabolism with nitrate or nitrous oxide as the terminal electron acceptor. The major fatty acids were $C_{16:0}$, $C_{18:1}\omega7c$ and summed feature 3 ($C_{16:1}\omega7c$ and/or $C_{15:0}$ iso 2-OH). The polar lipids consisted of phosphatidylethanolamine (56·6 %) and phosphatidylglycerol (43·4 %). The isoprenoid quinones were Q-8 (81·5 %), Q-9 (11·1 %) and Q-10 (7·4 %). The DNA G+C content was 50·0 mol%. A phylogenetic analysis based on 16S rRNA gene sequences showed that strain BD1^T formed a distinct lineage in the *Gammaproteobacteria* and that it exhibited the highest level of sequence similarity with species of the genera *Alteromonas* (92·8–93·7 %), *Aestuariibacter* (93·0 %), *Glaciecola* (90·4–92·7 %) and *Salinimonas* (91·8 %). Strain BD1^T was distinguishable from species of these genera by the presence of Q-9 and Q-10. Phenotypically, strain BD1^T was also distinguishable from species of these genera in that it did not require NaCl for growth and was capable of denitrification. On the basis of the polyphasic data from this study, the isolate represents a novel species within a novel genus, for which the name *Bowmanella denitrificans* gen. nov., sp. nov. is proposed. The type strain of *Bowmanella denitrificans* is BD1^T (=BCRC 17491^T=JCM 13378^T).

Alteromonas-like bacteria (*Alteromonas*-related bacteria) belonging to the class *Gammaproteobacteria* comprise a large group of marine, heterotrophic, Gram-negative rods that are mainly non-fermentative and polarly flagellated. A wide variety of families and genera have been classified as belonging to this bacterial group, including the *Alteromonadaceae* (*Alteromonas*, *Aestuariibacter*, *Glaciecola* and *Salinimonas*), the *Colwelliaceae* (*Colwellia* and *Thalassomonas*), the *Ferrimonadaceae* (*Ferrimonas*), the *Idiomarinaceae* (*Idiomarina* and *Pseudidiomarina*), the *Moritellaceae* (*Moritella*), the *Pseudoalteromonadaceae* (*Pseudoalteromonas* and *Algicola*), the *Psychromonadaceae* (*Psychromonas*), the *Shewanellaceae* (*Shewanella*) and *Agarivorans*, *Oceanimonas*, *Oceanisphaera* and *Zobellella* (Ivanova *et al.*, 2004; Jean *et al.*, 2006; Jeon *et al.*, 2005; Kurahashi & Yokota, 2004; Lin & Shieh, 2006; Yi *et al.*, 2004). Only some species in these families and genera are reported to be denitrifying bacteria, e.g. *Shewanella denitrificans*, *Shewanella sediminis*, *Shewanella decolorationis*, *Zobellella denitrificans* and *Zobellella taiwanensis* (Brettar *et al.*, 2002; Lin & Shieh, 2006; Xu *et al.*, 2005; Zhao *et al.*, 2005, 2006).

A non-fermentative denitrifying isolate has been recovered in our laboratory from a seawater sample collected in the shallow coastal region of An-Ping Harbour, Tainan, Taiwan, during a survey of the diversity of denitrifying bacteria. The polyphasic data obtained in this study indicated that this denitrifying isolate could be classified within a novel genus of the *Alteromonas*-like gammaproteobacteria.

The GenBank/EMBL/DDBJ accession number for the 16S rRNA gene sequence of strain BD1^T is DQ343294.

Fatty acid compositions of strain BD1^T and related strains, electron micrographs and growth curves are available as supplementary material in IJSEM Online.

Polypeptone/yeast extract/nitrate (PYN) broth medium was used for the enrichment cultivation of denitrifying bacteria in the seawater. This medium and others used in this study were described in our recent report (Lin & Shieh, 2006).

An-Ping Harbour is located in the south-west of Taiwan. Seawater samples were collected from the shallow coastal region of this harbour in the morning at low tide. Each sample was decimally diluted with sterile NaCl/Tris buffer (30 g NaCl and 0·24 g Tris base in 1 l deionized water, pH 8·0). A 1 ml aliquot of each dilution (10^1–10^3) was transferred to a rimless tube (16 mm × 10 cm) containing PYN broth medium (5 ml) into which an inverted Durham tube had been placed. All culture tubes were incubated aerobically at 25 °C in the dark for 3–7 days. Cultures that developed visible turbidity and produced gas (which accumulated in the Durham tubes) were streaked (one loopful) on polypeptone/yeast extract (PY) plate medium. Individual colonies appearing on each plate were picked off and purified by successive streaking on PY plates. Only isolates that exhibited growth and produced gas in PYN broth were maintained in PY stab medium and stored at 25 °C. One of the isolates, designated strain BD1^T, was used for the present study.

Growth and other phenotypic properties used for the physiological and morphological characterization of strain BD1^T were examined according to established procedures, as described previously (Lin & Shieh, 2006).

Cells grown in PY broth at 30 °C for 2 days were harvested by centrifugation. Polar lipids in the cells were extracted, purified and analysed by the methods described by Lin & Shieh (2006). Isoprenoid quinones were analysed as described by Hu *et al.* (2001), using HPLC apparatus equipped with a reversed-phase column (4·6 × 250 mm; Waters model $5C_{18}$-AR-II). A mixture of methanol and isopropyl ether (9 : 2, v/v) was used as the mobile phase, and quinones were detected at 270 nm. Fatty acids in whole cells grown on PY plate medium at 30 °C for 2 days were extracted, saponified and esterified; this was followed by GC analysis of the fatty acid methyl esters according to the instructions of the MIDI system (Sasser, 1997). This work, and DNA G+C content determination by HPLC analysis (Shieh & Liu, 1996), was performed at the Bioresources Center for Research and Collection, Food Industry Research and Development Institute, Taiwan.

Strain BD1^T was cultivated aerobically in PY broth at 30 °C in the dark for 2 days. The culture was centrifuged to harvest the cells. Extraction and purification of total genomic DNA from the cells and PCR amplification of 16S rRNA gene were performed according to the methods described by Lin & Shieh (2006). The sequencing reaction for the 16S rRNA gene, alignment and comparison of the resulting sequence with reference sequences available in the GenBank database, calculation of distance matrices for the aligned sequences and reconstruction of a phylogenetic tree by the neighbour-joining method were done as described by Shieh *et al.* (2004). Bootstrap confidence values (Felsenstein, 1985) were obtained using 1000 resamplings with the option of stepwise addition. Phylogenetic trees were also constructed by using the maximum-parsimony (Fitch, 1971) and maximum-likelihood (Felsenstein, 1981) methods.

An almost-complete 16S rRNA gene sequence of strain BD1^T was determined (1459 bp). It was aligned and compared with all bacterial sequences available in the GenBank database. A phylogenetic analysis based on 16S rRNA gene sequences showed that strain BD1^T was a member of the *Alteromonas*-like bacteria in the class *Gammaproteobacteria*. A neighbour-joining tree, showing the phylogenetic relationship between strain BD1^T and selected representatives of *Alteromonas*-like bacteria, is presented in Fig. 1; similar results were obtained using the maximum-parsimony and maximum-likelihood algorithms (not shown). The phylogenetic analyses showed that strain BD1^T formed a robust cluster at sequence similarity levels of 90·4–93·7 % (91–140 differences out of 1259–1459 nucleotide sites) with species of the genera *Alteromonas* (92·8–93·7 %), *Aestuariibacter* (93·0 %), *Glaciecola* (90·4–92·7 %) and *Salinimonas* (91·8 %) in the family *Alteromonadaceae*. No other bacterial species shared more than 90 % sequence similarity with the strain. The low levels of sequence similarity with other bacteria indicated that strain BD1^T could be assigned to a novel genus. Phylogenetically, the strain could be considered to represent a novel family, since it was clearly an outgroup with respect to species of the family *Alteromonadaceae*. Moreover, the 16S rRNA gene sequences of *Alteromonadaceae* species have been defined as possessing signature nucleotides, 737 (A), 770 (T) and 809 (A) (Ivanova *et al.*, 2004), whereas strain BD1^T has nucleotides G, C and G, respectively, at these sequence positions.

The polar lipids of strain BD1^T consisted of phosphatidylethanolamine (56·6 %) and phosphatidylglycerol (43·4 %). These two phospholipids have also been reported to be the major constituents of the polar lipids in some *Alteromonas* species, such as *Alteromonas addita* and *Alteromonas macleodii* (Ivanova *et al.*, 2000, 2005). Phosphatidic acid, bis-phosphatidic acid and lysophosphatidylethanolamine were detected as minor constituents of polar lipids in these species (Ivanova *et al.*, 2000, 2005). However, none of these phospholipids has been detected in strain BD1^T. The isoprenoid quinones of strain BD1^T consisted of Q-8 (81·5 %), Q-9 (11·1 %) and Q-10 (7·4 %). Q-8 was also found to be the predominant isoprenoid quinone in species of the genera *Alteromonas*, *Aestuariibacter* and *Salinimonas* (Ivanova *et al.*, 2000, 2005; Jeon *et al.*, 2005; Martínez-Checa *et al.*, 2005; Yi *et al.*, 2004; Yoon *et al.*, 2003, 2004), but these species have never been shown to contain Q-9 or Q-10. The data from the analysis of isoprenoid quinones supported the assignment of strain BD1^T to a novel genus. Strain BD1^T, like species belonging to the *Alteromonadaceae*, contained $C_{16:0}$ (21·7 %), $C_{18:1}\omega7c$ (19·4 %) and summed feature 3 ($C_{16:1}\omega7c$ and/or $C_{15:0}$ iso 2-OH) (24·1 %; the two fatty

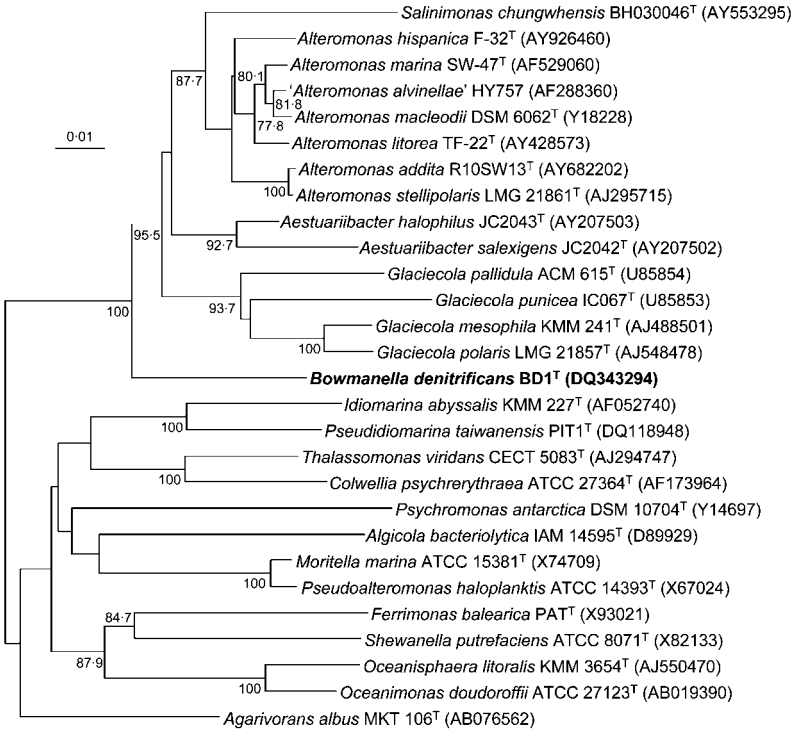

Fig. 1. Neighbour-joining dendrogram based on 16S rRNA gene sequences showing the phylogenetic relationship between strain BD1^T and species of the *Alteromonadaceae* and other families and genera in the *Alteromonas*-like gammaproteobacteria. GenBank accession numbers are given in parentheses. Bootstrap values (percentages of 1000 replicates) greater than 70 % are shown at branch points. Bar, 1 substitution per 100 nucleotides.

acids could not be differentiated in the MIDI system) as the major cellular fatty acids. Other cellular fatty acids present at levels greater than 3 % included $C_{16:0}$ N alcohol (3·7 %), $C_{17:1}\omega8c$ (3·8 %), $C_{12:0}$ 3-OH (7·1 %) and $C_{17:0}$ 10-methyl (3·5 %). Quantitative differences in the cellular fatty acids might serve to differentiate strain BD1^T from all species in the *Alteromonadaceae*, although the relevant species were not cultivated under the same conditions (see Supplementary Table S1 available in IJSEM Online). The DNA G+C content of strain BD1^T (50·0 mol%) fell within the range for *Aestuariibacter* (48·0–54·0 mol%) but was greater than those of *Alteromonas* (43·0–46·3 mol%), *Glaciecola* (40·0–46 mol%) and *Salinimonas* (48·0 mol%) (Table 1).

Strain BD1^T was distinguishable from species of the genera *Alteromonas*, *Aestuariibacter*, *Glaciecola* and *Salinimonas* in that it did not require NaCl for growth. The following combination of physiological characteristics also distinguished strain BD1^T from these species by at least three traits: the ability to reduce nitrate, the ability to denitrify, the production of indole, urea hydrolysis, growth at 10 and 40 °C and in 10 % NaCl and the inability to grow at 4 °C and in 12 % NaCl (Table 1). Other characteristics useful for differentiating strain BD1^T from species belonging to the family *Alteromonadaceae* are listed in Table 1.

The phylogenetic, chemotaxonomic and physiological data obtained in this study indicate that strain BD1^T should be assigned to a novel genus within the category of *Alteromonas*-like bacteria. Therefore the novel isolate represents a novel genus and species, for which the name *Bowmanella denitrificans* gen. nov., sp. nov. is proposed.

Description of *Bowmanella* gen. nov.

Bowmanella (Bow.man.el′la. L. dim ending *-ella*; N.L. fem. n. *Bowmanella* named after John P. Bowman, to honour his work in marine microbiology).

Members are Gram-negative rods belonging to the class *Gammaproteobacteria*. Cells grown in broth cultures are motile by means of a single, polar flagellum. Chemoorganotrophs capable of respiratory but not fermentative metabolism. Mesophilic, growing well at 25–35 °C; no growth occurs at 4 or 45 °C. NaCl stimulates growth but is not an absolute requirement. Oxidase- and catalase-positive. Major polar lipids are phosphatidylethanolamine and phosphatidylglycerol. Isoprenoid quinones consist of Q-8 (most abundant), Q-9 and Q-10. Major cellular fatty acids are $C_{16:0}$, $C_{18:1}\omega7c$ and summed feature 3 ($C_{16:1}\omega7c$ and/or $C_{15:0}$ iso 2-OH). The DNA G+C content of the type strain of the type species is 50·0 mol%. The type species is *Bowmanella denitrificans*.

Description of *Bowmanella denitrificans* sp. nov.

Bowmanella denitrificans (de.ni.tri′fi.cans. N.L. v. *denitrifico* to denitrify; N.L. part. adj. *denitrificans* denitrifying).

Description is as for the genus, with the following additional characteristics. Cells during late-exponential to early stationary phase of growth in broth cultures are polarly flagellated, curved rods (see Supplementary Fig. S1 in IJSEM Online), approximately 3·7–5·3 μm long by 1·2–1·6 μm wide. Cells form flake-like aggregations at late growth stages. Colonies produced on agar plates are circular,

Table 1. Characteristics useful for differentiating strain BD1^T from species of the genera *Alteromonas*, *Glaciecola*, *Aestuariibacter* and *Salinimonas* of the family *Alteromonadaceae*

Strains: 1, strain BD1^T; 2, *Alteromonas macleodii* DSM 6062^T (Baumann *et al.*, 1984; Yoon *et al.*, 2003); 3, *Alteromonas marina* KCCM 41638^T (Yoon *et al.*, 2003); 4, *Alteromonas litorea* TF-22^T (Yoon *et al.*, 2004); 5, *Alteromonas stellipolaris* LMG 21861^T (Van Trappen *et al.*, 2004a); 6, *Alteromonas addita* R10SW13^T (Ivanova *et al.*, 2005); 7, *Alteromonas hispanica* F-32^T (Martínez-Checa *et al.*, 2005); 8, *Glaciecola punicea* ACAM 611^T (Bowman *et al.*, 1998); 9, *Glaciecola pallidula* ACAM 615^T (Bowman *et al.*, 1998); 10, *Glaciecola polaris* LMG 21857^T (Van Trappen *et al.*, 2004b); 11, *Glaciecola mesophila* KMM 241^T (Romanenko *et al.*, 2003); 12, *Aestuariibacter salexigens* JC2942^T (unless indicated, data from Yi *et al.*, 2004); 13, *Aestuariibacter halophilus* JC2043^T (unless indicated, data from Yi *et al.*, 2004); 14, *Salinimonas chungwhensis* BH030046^T (Jeon *et al.*, 2005). Symbols: +, positive; −, negative; W, weakly positive; ND, no data available.

Characteristic	1	2	3	4	5	6	7	8	9	10	11	12	13	14
Nitrate reduction	+	−	−	−	−	−	−	−	−	−	−	+	+	−
Denitrification	+	−	−	−	−	−	−	−	−	−	−	−*	−*	−
Indole production	+	ND	−	ND	−	−	−	−	−	−	−	−	−	ND
Hydrolysis of:														
Casein	+	ND	+	+	ND	ND	+	−	−	−	−	−	+	+
Gelatin	+	+	+	+	+	W	+	−	−	−	−	+	+	+
Urea	+	ND	ND	−	−	ND	−	−	−	−	−	−	−	+
Utilization of:														
D-Fructose	+	+	+	+	+	+	−	−	−	+	+	−	−	−
D-Galactose	+	+	+	+	+	−	−	−	−	+	+	W	−	ND
Lactose	+	+	+	+	+	+	−	−	−	+	−	−	−	−
L-Lactate	−	+	−	−	−	+	−	−	+	−	−	ND	ND	ND
D-Mannitol	−	+	−	−	+	−	+	−	−	+	+	ND	ND	−
Acid production from D-glucose	−	+	−	−	−	−	−	W	W	−	W	−	−	+
Growth at/in:														
4 °C	−	−	−	+	−	+	+	+	+	−	−	−	−	−
10 °C	+	+	+	+	+	+	+	+	+	+	+	−	−	+
40 °C	+	+	+	+	−	−	+	−	−	−	−	+	+	+
0 % NaCl	+	−	−	−	−	−	−	−	−	−	−	−	−	−
10 % NaCl	+	−	+	+	+	+	+	−	−	+	−	+	+	+
12 % NaCl	−	−	+	+	−	−	+	−	−	−	−	−	−	+
Presence of Q-9 and Q-10	+	−	−	−	ND	ND	−	ND	ND	ND	ND	−	−	−
DNA G+C content (mol%)	50·0	46·0	45·0	46·0	43·3	43·4	46·3	43·9	40·0	44·0	44·8	48·0	54·0	48·0

*Data from this study.

off-white in colour and non-luminescent. Swarming does not occur. Growth occurs at temperatures between 10 and 40 °C, with an optimum at 30–35 °C; no growth occurs at 43 °C. Growth occurs at pH values in the range 6–10, with an optimum at pH 8. Growth occurs at NaCl levels of 0–10 %, with an optimum at 1–3 %; no growth occurs at 12 % NaCl. Unable to ferment D-glucose, D-arabinose, L-arabinose, D-cellobiose, D-galactose, D-mannose, sucrose, D-trehalose, D-xylose, dulcitol, inositol or D-mannitol. Molecular oxygen is a universal electron acceptor while anaerobic growth can be achieved by carrying out denitrification with NO_3^- or N_2O as the terminal electron acceptor (see Supplementary Fig. S2 in IJSEM Online). Indole is produced from tryptophan. H_2S is not produced from thiosulfate. Aesculin, casein, DNA, gelatin, lecithin, starch, Tween 80 and urea are hydrolysed, but agar is not. Arginine dihydrolase, lysine decarboxylase and ornithine decarboxylase activities are not detected. D-Cellobiose, D-fructose, D-glucose, D-galactose, D-lactose, D-mannose, sucrose, D-trehalose, D-xylose, acetate and β-hydroxybutyrate can be utilized as sole carbon and energy sources, but D-arabinose, L-arabinose, adonitol, dulcitol, inositol, D-mannitol, D-melezitose, D-melibiose, D-sorbitol and L-lactate cannot. The following constitutive enzyme activities are detected in API ZYM tests: leucine arylamidase, valine arylamidase, esterase (C4), esterase lipase (C8), *N*-acetyl-β-glucosaminidase, naphthol-AS-BI-phosphohydrolase, acid phosphatase and alkaline phosphatase. Resistant to the vibriostatic agent O/129 at 10–150 μg. Susceptible to chloramphenicol (30 μg), colistin (10 μg), nalidixic acid (30 μg), neomycin (30 μg), polymyxin B (300 U) and tetracycline (30 μg) and intermediately susceptible to gentamicin (10 μg) and kanamycin (30 μg). Resistant to ampicillin (10 μg), carbenicillin (100 μg), cephalothin (30 μg), clindamycin (2 μg), erythromycin (15 μg), lincomycin (2 μg), novobiocin (30 μg), oxacillin (1 μg), penicillin G (10 μg), streptomycin (10 μg) and vancomycin (30 μg). Polar lipids consist of phosphatidylethanolamine (56·6 %) and phosphatidylglycerol (43·4 %).

Isoprenoid quinones consist of Q-8 (81·5 %), Q-9 (11·1 %) and Q-10 (7·4 %). Cellular fatty acids consist of $C_{16:0}$ (21·7 %), $C_{18:1}\omega7c$ (19·4 %), summed feature 3 ($C_{16:1}\omega7c$ and/or $C_{15:0}$ iso 2-OH) (24·1 %), $C_{12:0}$ 3-OH (7·1 %), $C_{17:1}\omega8c$ (3·8 %), $C_{16:0}$ N alcohol (3·7 %), $C_{17:0}$ 10-methyl (3·5 %), $C_{12:0}$ (2·5 %), $C_{14:0}$ (2·1 %), $C_{16:1}\omega7c$ alcohol (2·1 %), $C_{17:0}$ (1·6 %) $C_{18:0}$ (1·5 %), $C_{19:0}$ 10-methyl (1·2 %), $C_{11:0}$ 3-OH (1·0 %), $C_{13:1}$ (1·0 %), $C_{10:0}$ 3-OH (0·8 %), $C_{16:0}$ 3-OH (0·8 %) and $C_{10:0}$ (0·6 %).

The type strain, BD1^T (= BCRC 17491^T = JCM 13378^T), was isolated from shallow coastal water from An-Ping Harbour, Tainan, Taiwan.

Acknowledgements

We are grateful to Associate Editor Dr Hans-Jürgen Busse and two anonymous referees for valuable comments. We are also grateful to Dr Jongsik Chun (School of Biological Sciences, Seoul National University, Republic of Korea) for supplying the type strains of *Aestuariibacter salexigens* and *Aestuariibacter halophilus*. This study was supported by grants NSC91-2313-B-002-327, NSC92-2313-B-002-084, NSC92-3114-B-002-014 and NSC92-2211-E-426-008 from the National Science Council (Taiwan).

References

Baumann, P., Baumann, L., Bowditch, R. D. & Beaman, B. (1984). Taxonomy of *Alteromonas*: *A. nigrifaciens* sp. nov., nom. rev.; *A. macleodii* and *A. haloplanktis*. *Int J Syst Bacteriol* **34**, 145–149.

Bowman, J. P., McCammon, S. A., Brown, J. L. & McMeekin, T. A. (1998). *Glaciecola punicea* gen. nov., sp. nov. and *Glaciecola pallidula* gen. nov., sp. nov.: psychrophilic bacteria from Antarctic sea-ice habitats. *Int J Syst Bacteriol* **48**, 1213–1222.

Brettar, I., Christen, R. & Höfle, M. G. (2002). *Shewanella denitrificans* sp. nov., a vigorously denitrifying bacterium isolated from the oxic–anoxic interface of the Gotland Deep in the central Baltic Sea. *Int J Syst Evol Microbiol* **52**, 2211–2217.

Felsenstein, J. (1981). Evolutionary trees from DNA sequences: a maximum likelihood approach. *J Mol Evol* **17**, 368–376.

Felsenstein, J. (1985). Confidence limits on phylogenies: an approach using the bootstrap. *Evolution* **35**, 22–33.

Fitch, W. M. (1971). Toward defining the course of evolution: minimum change for a specific tree topology. *Syst Zool* **20**, 406–416.

Hu, H.-Y., Lim, B.-R., Goto, N. & Fujie, K. (2001). Analytical precision and repeatability of respiratory quinones for quantitative study of microbial community structure in environmental samples. *J Microbiol Methods* **47**, 17–24.

Ivanova, E. P., Zhukova, N. V., Svetashev, V. I., Gorshkova, N. M., Kurilenko, V. V., Frolova, G. M. & Mikhailov, V. V. (2000). Evaluation of phospholipid and fatty acid compositions as chemotaxonomic markers of *Alteromonas*-like Proteobacteria. *Curr Microbiol* **41**, 341–345.

Ivanova, E. P., Flavier, S. & Christen, R. (2004). Phylogenetic relationships among marine *Alteromonas*-like proteobacteria: emended description of the family *Alteromonadaceae* and proposal of *Pseudoalteromonadaceae* fam. nov., *Colwelliaceae* fam. nov., *Shewanellaceae* fam. nov., *Moritellaceae* fam. nov., *Ferrimonadaceae* fam. nov., *Idiomarinaceae* fam. nov. and *Psychromonadaceae* fam. nov. *Int J Syst Evol Microbiol* **54**, 1773–1788.

Ivanova, E. P., Bowman, J. P., Lysenko, A. M., Zhukova, N. V., Gorshkova, N. M., Sergeev, A. F. & Mikhailov, V. V. (2005). *Alteromonas addita* sp. nov. *Int J Syst Evol Microbiol* **55**, 1065–1068.

Jean, W. D., Shieh, W. Y. & Chiu, H.-H. (2006). *Pseudidiomarina taiwanensis* gen. nov., sp. nov., a marine bacterium isolated from shallow coastal water of An-Ping Harbour, Taiwan, and emended description of the family *Idiomarinaceae*. *Int J Syst Evol Microbiol* **56**, 899–905.

Jeon, C. K., Lim, J.-M., Park, D.-J. & Kim, C.-J. (2005). *Salinimonas chungwhensis* gen. nov., sp. nov., a moderately halophilic bacterium from a solar saltern in Korea. *Int J Syst Evol Microbiol* **55**, 239–243.

Kurahashi, M. & Yokota, A. (2004). *Agarivorans albus* gen. nov., sp. nov., a γ-proteobacterium isolated from marine animals. *Int J Syst Evol Microbiol* **54**, 693–697.

Lin, Y.-T. & Shieh, W. Y. (2006). *Zobellella denitrificans* gen. nov., sp. nov. and *Zobellella taiwanensis* sp. nov., two denitrifying bacteria capable of fermentative metabolism. *Int J Syst Evol Microbiol* **56**, 1209–1215.

Martínez-Checa, F., Bejar, V., Llamas, I., del Moral, A. & Quesada, E. (2005). *Alteromonas hispanica* sp. nov., a polyunsaturated-fatty-acid-producing, halophilic bacterium isolated from Fuente de Piedra, southern Spain. *Int J Syst Evol Microbiol* **55**, 2385–2390.

Romanenko, L. A., Zhukova, N. V., Rohde, M., Lysenko, A. M., Mikhailov, V. V. & Stackebrandt, E. (2003). *Glaciecola mesophila* sp. nov., a novel marine agar-digesting bacterium. *Int J Syst Evol Microbiol* **53**, 647–651.

Sasser, M. (1997). *Identification of bacteria by gas chromatography of cellular fatty acids*. MIDI Technical Note 101. Newark, DE: MIDI.

Shieh, W. Y. & Liu, C. M. (1996). Denitrification by a novel halophilic fermentative bacterium. *Can J Microbiol* **42**, 507–514.

Shieh, W. Y., Lin, Y.-T. & Jean, W. D. (2004). *Pseudovibrio denitrificans* gen. nov., sp. nov., a marine, facultatively anaerobic, fermentative bacterium capable of denitrification. *Int J Syst Evol Microbiol* **54**, 2307–2312.

Van Trappen, S., Tan, T.-L., Yang, J., Mergaert, J. & Swings, J. (2004a). *Alteromonas stellipolaris* sp. nov., a novel, budding, prosthecate bacterium from Antarctic seas, and emended description of the genus *Alteromonas*. *Int J Syst Evol Microbiol* **54**, 1157–1163.

Van Trappen, S., Tan, T.-L., Yang, J., Mergaert, J. & Swings, J. (2004b). *Glaciecola polaris* sp. nov., a novel budding and prosthecate bacterium from the Arctic Ocean, and emended description of the genus *Glaciecola*. *Int J Syst Evol Microbiol* **54**, 1765–1771.

Xu, M., Guo, J., Cen, Y., Zhong, X., Cao, W. & Sun, G. (2005). *Shewanella decolorationis* sp. nov., a dye-decolorizing bacterium isolated from activated sludge of a waste-water treatment plant. *Int J Syst Evol Microbiol* **55**, 363–368.

Yi, H., Bae, K. S. & Chun, J. (2004). *Aestuariibacter salexigens* gen. nov., sp. nov. and *Aestuariibacter halophilus* sp. nov., isolated from tidal flat sediment, and emended description of *Alteromonas macleodii*. *Int J Syst Evol Microbiol* **54**, 571–576.

Yoon, J.-H., Kim, I.-G., Kang, K. H., Oh, T.-K. & Park, Y.-H. (2003). *Alteromonas marina* sp. nov., isolated from sea water of the East Sea in Korea. *Int J Syst Evol Microbiol* **53**, 1625–1630.

Yoon, J.-H., Yeo, S.-H., Oh, T.-K. & Park, Y.-H. (2004). *Alteromonas litorea* sp. nov., a slightly halophilic bacterium isolated from an intertidal sediment of the Yellow Sea in Korea. *Int J Syst Evol Microbiol* **54**, 1197–1201.

Zhao, J.-S., Manno, D., Beaulieu, C., Paquet, L. & Hawari, J. (2005). *Shewanella sediminis* sp. nov., a novel Na^+-requiring and hexahydro-1,3,5-trinitro-1,3,5-triazine-degrading bacterium from marine sediment. *Int J Syst Evol Microbiol* **55**, 1511–1520.

Zhao, J.-S., Manno, D., Leggiadro, C., O'Neil, D. & Hawari, J. (2006). *Shewanella halifaxensis* sp. nov., a novel obligately respiratory and denitrifying psychrophile. *Int J Syst Evol Microbiol* **56**, 205–212.