Exiguobacterium soli sp. nov., a psychrophilic bacterium from the McMurdo Dry Valleys, Antarctica

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A novel psychrophilic bacterium, designated strain DVS 3Y^T, was isolated from a moraine sample from the McMurdo Dry Valleys, Antarctica. Phenotypic and chemotaxonomic characteristics and data from a phylogenetic analysis based on 16S rRNA gene sequences indicated that strain DVS 3Y^T was related to the genus *Exiguobacterium*. Strain DVS 3Y^T exhibited >97 % gene sequence similarity with respect to *Exiguobacterium acetylicum* DSM 20416^T (97.4 %), *Exiguobacterium oxidotolerans* DSM 17272^T (97.4 %), *Exiguobacterium indicum* IAM 15368^T (97.6 %), *Exiguobacterium undae* DSM 14481^T (98.2 %), *Exiguobacterium sibiricum* DSM 17290^T (98.6 %) and *Exiguobacterium antarcticum* DSM 14480^T (99.6 %). However, the levels of DNA–DNA relatedness between strain DVS 3Y^T and *E. acetylicum* DSM 20416^T, *E. oxidotolerans* JCM 12280^T, *E. indicum* IAM 15368^T, *E. undae* DSM 14481^T, *E. sibiricum* DSM 17290^T and *E. antarcticum* DSM 14480^T were less than 70 % (52, 25, 16, 33, 16 and 38 %, respectively). Strain DVS 3Y^T also differed from these six closely related species in a number of phenotypic traits. Phenotypic, chemotaxonomic and phylogenetic data suggest that strain DVS 3Y^T represents a novel species of the genus *Exiguobacterium*, for which the name *Exiguobacterium soli* sp. nov. is proposed. The type strain is DVS 3Y^T (=MTCC 4816^T=JCM 14376^T).

The genus *Exiguobacterium* was created by Collins *et al.* (1983) to accommodate a Gram-positive, non-spore-forming, facultatively anaerobic, alkaliphilic bacterium isolated from potato-processing effluent (Gee *et al.*, 1980). At the time of writing, 11 species had been described in this genus: *Exiguobacterium aurantiacum* (Collins *et al.*, 1983), *E. undae* and *E. antarcticum* (Fruhling *et al.*, 2002), *E. oxidotolerans* (Yumoto *et al.*, 2004), *E. aestuarii* and *E. marinum* (Kim *et al.*, 2005), *E. mexicanum* and *E. artemiae* (Lopez-Cortes *et al.*, 2006), *E. acetylicum* (which was transferred from the genus *Brevibacterium*; Farrow *et al.*, 1994), *E. indicum* (Chaturvedi & Shivaji, 2006) and *E. sibiricum* (Rodrigues *et al.*, 2006). In the present study, strain DVS 3Y^T, a Gram-positive, motile, non-spore-forming, rod-shaped bacterium isolated from a moraine sample from Antarctica, was identified as a novel species of the genus *Exiguobacterium*.

Strain DVS 3Y^T was isolated from a moraine sample collected in December 1995 from the McMurdo Dry

Valleys in Antarctica. The sample was transported in ice and stored at –20 °C until used for analysis. Following suspension in sterile saline (150 mM), the sample was serially diluted, plated on YPD agar containing yeast extract (0.5 %), peptone (1 %), glucose (2 %) and agar (1.5 %) and incubated at 25 °C for 4 days to determine the bacterial density. The sample yielded about 4.5–5 × 10⁴ c.f.u. ml^{–1}. Colonies of strain DVS 3Y^T, which were yellowish orange, were purified by means of repeated streaking and were maintained on ABM agar (pH 7.2) containing yeast extract (1.5 %), peptone (0.2 %) and agar (1.5 %) and incubated at 22 °C. ABM agar was also used for the determination of growth at various temperatures (2.5, 15, 22, 28, 37 and 40 °C), pH values (4, 6, 7, 8, 10 and 11) and NaCl concentrations (5, 10 and 15 %) (Shivaji *et al.*, 1989). The buffers used were MOPS (for pH 6.5–7.9) and CAPS (for pH 9.7–11.1). Phenotypic characteristics such as colony morphology, cell morphology, motility, various enzyme activities (as listed in Table 1) and sensitivity to antibiotics at 22 °C were ascertained using standard methods (Lányi, 1987; Smibert & Krieg, 1994). Gas production was determined according to the method of Hugh & Leifson (1953). For the determination of acid production, the medium used was phenol red agar base (pH 7.4) containing protease peptone (1 %), beef extract

The GenBank/EMBL/DDBJ accession number for the 16S rRNA gene sequence of strain DVS 3Y^T is AY864633.

The fatty acid compositions of strain DVS 3Y^T and related species of the genus *Exiguobacterium* are compared in a supplementary table available with the online version of this paper.

Table 1. Phenotypic characteristics that serve to differentiate strain DVS 3Y^T from the six most closely related species of the genus *Exiguobacterium*

Strains: 1, DVS 3Y^T; 2, *E. antarcticum* DSM 14480^T; 3, *E. sibiricum* DSM 17290^T; 4, *E. undae* DSM 14481^T; 5, *E. indicum* IAM 15368^T; 6, *E. oxidotolerans* JCM 12280^T; 7, *E. acetylicum* DSM 20416^T. All data are from this study unless otherwise indicated. All strains are motile, Gram-positive, rod-shaped, do not form endospores, grow at between 10 and 30 °C (with an optimum at 25 °C), tolerate up to 5.8% NaCl, grow in peptone, fail to grow at pH 4, grow at pH 6–10, are positive in tests for catalase, oxidase, β -galactosidase, arginine dihydrolase and ornithine decarboxylase and for the Voges–Proskauer test, but are negative for lipase, urease and H₂S production and for the indole test. All taxa utilize D-glucose, sucrose, glycerol, dextrin and thioglycolate, but not sorbose, N-acetylglucosamine, polyethylene glycol, D-adonitol, L-phenylalanine, hydroxybutyric acid or sodium propionate. All are negative for acid production from D-sorbitol, D-lactose and L-arabinose. All are sensitive to the following antibiotics (μ g): tobramycin (15), lomefloxacin (30), roxithromycin (30), ciprofloxacin (30), lincomycin (15), cefotaxime (30), cefazolin (30), kanamycin (30), novobiocin (30), chloramphenicol (30), ampicillin (25), tetracycline (30), streptomycin (25), erythromycin (15), bacitracin (10), gentamicin G (30), polymyxin B (50), oleandomycin (15), spectinomycin (100), rifampicin (25) and carbenicillin (100). +, Positive; –, negative; ND, not determined; w, weakly positive; R, resistant; S, sensitive; DPG, diphosphatidylglycerol; PE, phosphatidylethanolamine; PG, phosphatidylglycerol; PI, phosphatidylinositol; PS, phosphatidylserine.

Characteristic	1	2	3	4	5	6	7
Habitat	Moraine sample from Antarctica	Microbial mat from a lake in Antarctica	Siberian permafrost	Garden pond	Glacial water	Drainage from fish-processing plant	Creamery waste
Colony size (mm)	2–3	2–3	2–3	2–4	2–4	1–5	2–5
Colony shape	Round	Round	Round	Round	Round	Round	Irregular
Colony colour	Yellowish orange	Orange	Orange	Orange	Yellowish orange	Orange	Yellowish orange
Growth temperature (°C)							
2.5	+	–	+	–	w	w	–
5	+	–	+	–	+	+	–
37	–	+	+	+	–	+	w
Maximum growth temperature (°C)	30	41	40	41	30	40	37
Gelatinase	+	+	+	+	–	+	+
DNase	–	+	–	+	–	+	+
Caseinase	+	+	+	+	–	+	+
Phosphatase	+	+	+	–	+	+	+
Lysine decarboxylase	+	–	+	+	+	–	–
Arginine decarboxylase	+	+	+	+	+	–	–
Tryptophan deamination	–	+	+	–	–	+	+
Aesculin hydrolysis	+	+	+	+	–	+	+
Starch hydrolysis	+	+	+	+	–	+	+
Citrate utilization	–	–	+	–	+	+	+
Malonate utilization	–	–	–	–	+	–	–
Reduction of nitrate to nitrite	–	–	–	–	+	–	–
Methyl red test	–	+	–	+	–	+	+
Acid production from:							
Melibiose	–	–	+	–	–	–	–
D-Adonitol	–	–	–	–	–	–	+
Cellobiose	+	+	–	–	–	–	+
D-Fructose	+	+	+	+	–	+	+
D-Glucose	+	+	+	+	–	+	+
D-Mannose	+	–	+	+	–	+	+
Maltose	+	+	+	+	–	+	+
D-Ribose	+	+	+	+	–	+	+
L-Rhamnose	+	+	–	+	–	+	+
Sucrose	+	+	+	+	–	+	+
Trehalose	+	+	+	+	–	+	+
L-Xylose	+	–	+	–	–	–	–
Inulin	+	–	+	–	–	–	–
Carbon source utilization							
L-Arabinose	+	–	+	–	–	–	–
D-Arabinose	+	–	+	–	–	–	–
D-Ribose	+	+	+	+	–	+	+

Table 1. cont.

Characteristic	1	2	3	4	5	6	7
D-Xylose	+	—	—	—	+	—	—
L-Xylose	+	—	—	—	+	—	—
D-Fructose	+	+	+	+	—	+	+
D-Galactose	+	—	—	+	+	—	—
D-Mannose	+	+	—	+	—	+	+
L-Rhamnose	+	—	—	—	+	—	—
Cellobiose	+	+	—	+	+	—	+
D-Lactose	+	—	+	—	—	—	—
Maltose	+	+	—	+	—	+	+
L-Fucose	—	—	—	—	—	—	+
Melibiose	+	—	+	—	+	—	—
Trehalose	+	+	+	+	—	+	+
Melezitose	+	—	—	—	—	—	+
Raffinose	+	+	—	+	+	—	—
Cellulose	—	—	—	+	—	—	—
Inulin	+	—	—	—	+	—	—
Starch	+	+	+	+	—	+	+
Amygdalin	+	+	+	+	—	+	—
Arbutin	+	+	—	+	+	+	—
Salicin	—	—	ND	+	—	—	+
Glycogen	+	+	—	+	+	—	—
myo-Inositol	+	—	—	—	+	—	—
Dulcitol	+	—	—	—	+	—	—
Erythritol	+	—	ND	+	+	+	—
D-Mannitol	—	—	—	+	—	+	+
D-Sorbitol	+	—	+	—	+	—	—
Xylitol	—	—	—	—	—	—	+
Methyl α -D-mannoside	+	+	—	—	—	—	+
Methyl α -D-glucoside	—	—	—	+	—	+	+
Methyl α -D-galactoside	—	—	—	—	+	—	—
Methyl β -D-galactoside	—	—	—	—	+	—	—
α -Ketoglutaric acid	+	—	—	+	+	—	—
Citric acid	—	+	—	+	+	—	—
Fumaric acid	—	—	—	—	+	—	—
γ -Glucuronic acid	+	—	—	+	+	—	—
Lactic acid	+	—	—	—	+	—	—
L-Malic acid	—	—	—	—	+	—	—
Valeric acid	—	+	ND	—	+	—	—
Sodium acetate	—	+	—	+	+	—	—
5-Ketogluconate	w	—	—	—	+	—	—
Sodium gluconate	—	—	ND	—	+	—	—
Sodium fumarate	—	+	ND	—	+	—	—
Sodium formate	—	+	ND	—	+	—	—
Sodium succinate	—	—	ND	+	+	—	—
Potassium acetate	—	—	—	+	+	—	—
Sodium pyruvate	+	+	—	+	—	+	+
Amino acid utilization							
L-Alanine	+	+	+	+	+	—	—
L-Arginine	+	+	+	+	+	—	—
L-Aspartic acid	—	—	—	+	+	+	—
L-Asparagine	+	+	—	+	+	—	—
L-Cysteine	—	+	—	+	—	—	—
L-Creatinine	+	—	—	—	+	—	—
L-Glycine	+	—	—	+	+	—	—
L-Glutamine	+	+	+	+	+	+	—
L-Glutamic acid	+	+	+	—	+	—	—
L-Histidine	+	+	+	+	+	—	—

Table 1. cont.

Characteristic	1	2	3	4	5	6	7
L-Isoleucine	+	+	—	+	+	+	—
L-Leucine	+	+	—	+	+	—	+
L-Lysine	+	+	+	+	+	+	—
L-Methionine	—	+	+	+	+	+	—
L-Ornithine	+	+	—	+	+	—	—
L-Serine	—	—	+	+	+	+	—
L-Threonine	—	+	+	+	+	—	—
L-Proline	+	+	+	+	+	—	—
L-Tryptophan	—	+	ND	—	+	—	—
L-Tyrosine	—	+	ND	+	+	—	+
L-Valine	—	+	ND	+	+	—	—
Antibiotic tolerance (µg per disc)							
Amikacin (30)	S	S	S	S	R	S	S
Amoxicillin (30)	R	S	S	S	R	S	S
Clindamycin (25)	S	S	S	S	R	S	R
Colistin (10)	R	R	R	S	R	R	S
Doxycycline (25)	R	S	R	S	R	S	S
Co-trimoxazole (25)	S	S	S	S	R	S	S
Nalidixic acid (30)	S	S	S	S	R	S	S
Norfloxacin (10)	R	S	R	S	R	S	S
Nitrofurantoin (300)	R	S	S	S	R	S	S
Sulfamethoxazole (50)	R	S	R	R	R	S	S
DNA G + C content (mol%)	48.5	47	47.7*	48	48	47	47
Peptidoglycan†	Lys–Gly	ND	Lys–Gly	Lys–Asp	Lys–Gly	Lys–Gly	Lys–Gly
Quinone(s)	MK-6, MK-7, MK-8	MK-6, MK-7, MK-8	MK-6, MK-7, MK-8	MK-6, MK-7, MK-8	MK-7, MK-8	MK-6, MK-7, MK-8	MK-7
Polar lipids	PG, DPG, PE, PS, PI, unidentified	PG, DPG, PE, PS, PI, unidentified	PG, DPG, PE	PG, DPG, PE, PS, PI, unidentified	PG, DPG, PE (tr)	PG, DPG, PE, PS, PI, unidentified	DPG, PS

*Data from Rodrigues *et al.* (2006).

†Peptidoglycan determinations for strain DVS 3Y^T, *E. undae* DSM 14481^T, *E. indicum* IAM 15368^T, *E. acetylicum* DSM 20416^T are from this study; those for *E. antarcticum* DSM 14480^T, *E. sibiricum* DSM 17290^T and *E. oxidotolerans* JCM 12280^T are from Fruhling *et al.* (2002), Rodrigues *et al.* (2006) and Yumoto *et al.* (2004).

(0.1 %), NaCl (0.5 %), phenol red (0.0025 %) and agar (1.8 %). Minimal medium [K₂HPO₄, 1.05 % (w/v); KH₂PO₄, 0.45 % (w/v); (NH₄)₂SO₄, 0.1 % (w/v); agar, 1.5 % (w/v)] was used to evaluate the ability of the culture to assimilate various carbon compounds including amino acids (0.5 %, w/v) when provided as sole carbon sources. Fatty acid methyl esters were prepared according to the method of Sato & Murata (1988) from cells grown on tryptic soy medium (M322; Himedia) at 30 °C and analysed using the Microbial Identification System (MIDI) according to the protocol described by Agilent Technologies. The G + C content of the DNA was determined according to the spectrophotometric method (Shivaji *et al.*, 1989, 2004, 2005). Isoprenoid quinones were extracted following the protocol described by Collins *et al.* (1977), separated using HPLC and identified as described by Reddy *et al.* (2003). Peptidoglycan was prepared and analysed according to the method described by Komagata & Suzuki (1987). DNA–DNA hybridization was performed by using the membrane filter method (Tourova &

Antonov, 1987), as described previously (Shivaji *et al.*, 1992). *E. acetylicum* DSM 20416^T, *E. oxidotolerans* JCM 12280^T, *E. indicum* IAM 15368^T, *E. sibiricum* DSM 17290^T, *E. undae* DSM 14481^T and *E. antarcticum* DSM 14480^T were used as reference strains in studies relating to morphology, biochemical tests and the identification of fatty acids.

To establish the phylogenetic position of strain DVS 3Y^T, DNA was purified and the 16S rRNA gene was amplified and sequenced as described previously (Shivaji *et al.*, 2000; Chaturvedi *et al.*, 2005). The almost-complete sequence of 1518 nt was manually aligned against closely related species by using CLUSTAL W (Thompson *et al.*, 1994). Pairwise evolutionary distances were computed using the DNADIST program with the Kimura two-parameter model, as developed by Kimura (1980). Phylogenetic trees were constructed using the UPGMA and neighbour-joining tree-making algorithms in the PHYLIP package (Felsenstein, 1993). Stability among the clades of the phylogenetic tree

Fig. 1. Neighbour-joining tree based on 16S rRNA gene sequences showing the phylogenetic relationship between strain DVS 3Y^T and species of the genus *Exiguobacterium*. *Bacillus indicus* Sd/3^T was used as the outgroup. Bootstrap values greater than 50% are given at the nodes. Bar, 1 substitution per 100 nucleotides.

was assessed by taking 1000 replicates and analysing the dataset using the SEQBOOT, DNADIST, NEIGHBOR and CONSENSE programs in the PHYLIP package.

Phenotypic and chemotaxonomic characteristics of strain DVS 3Y^T are given in the species description, in Table 1 and in Supplementary Table S1 (available with IJSEM Online). Strain DVS 3Y^T was found to comprise cells that were Gram-positive, motile, rod-shaped and non-spore-forming, to possess MK-6, MK-7 and MK-8 as the major menaquinones, to possess cell-wall peptidoglycan of the Lys–Gly type and to have a DNA G+C content of 48.5 mol%; and it was therefore similar to recognized species of the genus *Exiguobacterium* (Collins *et al.*, 1983; Farrow *et al.*, 1994; Fruhling *et al.*, 2002; Yumoto *et al.*, 2004; Kim *et al.*, 2005; Lopez-Cortes *et al.*, 2006). Phylogenetic analysis based on the 16S rRNA gene sequence (1518 nt) further confirmed the affiliation of strain DVS 3Y^T to the genus *Exiguobacterium* (Fig. 1). Strain DVS 3Y^T formed a robust cluster, with a bootstrap value >74% with respect to *E. antarcticum* DSM 14480^T, *E. undae* DSM 14481^T, *E. artemiae* DSM 16484^T, *E. sibiricum* DSM 17290^T, *E. oxidotolerans* JCM 12280^T, *E. indicum* HHS 31^T and *E. acetylicum* DSM 20416^T (Fig. 1). The remaining strains, *E. aestuarii* DSM 16306^T, *E. marinum* DSM 16307^T, *E. aurantiacum* NCDO 2321^T and *E. mexicanum* DSM 16483^T, formed another clade. The formation of two distinct clades by species of the genus *Exiguobacterium* has been reported previously (Lopez-Cortes *et al.*, 2006; Kim *et al.*, 2005; Chaturvedi & Shivaji, 2006).

Using GeneTool 1.0 (http://www.biotoools.com/downloads/content/up_con.html), the 16S rRNA gene sequence similarity of strain DVS 3Y^T with the recognized species of the genus *Exiguobacterium* was as follows: 92.5% with *E. aestuarii* TF-16^T (GenBank accession no. AY594264), 92.5% with *E. marinum* TF-80^T (AY594266), 93.1% with *E. aurantiacum* NCDO 2321^T (X70316), 93.6% with *E.*

mexicanum DSM 16483^T (AM072764), 96.8% with *E. artemiae* DSM 16484^T (AM072763), 97.4% with *E. acetylicum* DSM 20416^T (D55730), 97.4% with *E. oxidotolerans* DSM 17272^T (AB105164), 97.6% with *E. indicum* IAM 15368^T (AJ846291), 98.2% with *E. undae* DSM 14481^T (AJ344151), 98.5% with *E. sibiricum* DSM 17290^T (NZ_AADW02000023) and 99.6% with *E. antarcticum* DSM 14480^T (AJ297437). Thus, to merit the status of a novel species, strain DVS 3Y^T must be differentiated from *E. acetylicum* DSM 20416^T, *E. oxidotolerans* DSM 17272^T, *E. indicum* IAM 15368^T, *E. undae* DSM 14481^T, *E. sibiricum* DSM 17290^T and *E. antarcticum* DSM 14480^T with which it exhibits >97% similarity at the 16S rRNA gene level.

At the whole-genome level, as determined by DNA–DNA hybridization, when strain DVS 3Y^T was radioactively labelled, the levels of DNA–DNA relatedness between strain DVS 3Y^T and *E. acetylicum* DSM 20416^T, *E. oxidotolerans* JCM 12280^T, *E. indicum* IAM 15368^T, *E. undae* DSM 14481^T, *E. sibiricum* DSM 17290^T and *E. antarcticum* DSM 14480^T were 52, 25, 16, 33, 16 and 38%, respectively. However, when *E. acetylicum* DSM 20416^T, *E. oxidotolerans* JCM 12280^T, *E. indicum* IAM 15368^T, *E. undae* DSM 14481^T, *E. sibiricum* DSM 17290^T and *E. antarcticum* DSM 14480^T were labelled and used for DNA–DNA hybridization with DVS 3Y^T in the reciprocal reaction, the similarities were 42, 23, 23, 39, 24 and 33%, respectively.

Strain DVS 3Y^T differed from the above six phylogenetically closely related species in terms of a number of phenotypic characteristics (Table 1) and also exhibited significant quantitative differences with regard to fatty acid content (see Supplementary Table S1 in IJSEM Online). The predominant fatty acids were iso-C_{13:0} (16.1%), iso-C_{15:0} (17.1%), iso-C_{16:0} (4.1%), iso-C_{17:0} (10.6%), anteiso-C_{13:0} (15.2%), anteiso-C_{15:0} (6.2%), anteiso-C_{17:0} (3.3%) and C_{16:0} (3.6%). These iso- and anteiso-fatty acids constituted >63% of the total fatty acids in

strain DVS 3Y^T, *E. sibiricum* DSM 17290^T, *E. undae* DSM 14481^T, *E. indicum* IAM 15368^T and *E. oxidotolerans* JCM 12280^T. These *Exiguobacterium* species (with the exception of *E. undae* DSM 14481^T) grow at 5 °C, unlike the other species. Thus, these iso- and anteiso- fatty acids may be required for growth at low temperatures. According to the criteria generally used for discriminating species (Stackebrandt & Goebel, 1994), strain DVS 3Y^T, which exhibits <70% similarity at the DNA–DNA level with respect to *E. acetylicum* DSM 20416^T, *E. oxidotolerans* JCM 12280^T, *E. indicum* IAM 15368^T, *E. undae* DSM 14481^T, *E. sibiricum* DSM 17290^T and *E. antarcticum* DSM 14480^T (the most closely related species) and which also differs phenotypically from these six species (Table 1 and Supplementary Table S1), represents a novel species in the genus *Exiguobacterium*. The name *Exiguobacterium soli* sp. nov. is proposed for this novel species.

Description of *Exiguobacterium soli* sp. nov.

Exiguobacterium soli sp. nov. (so'li. L. gen. n. soli of soil).

Cells are aerobic, Gram-positive, motile and rod-shaped (1.9 µm in length and 0.95 µm in width). Stationary-phase cells are coccobacillary in shape (1.5 µm in length and 1.15 µm in diameter). Colonies (2–3 mm) on nutrient agar are round, shiny, irregular, elevated and yellowish orange-coloured after 24 h at 22 °C. No spores are observed. Growth occurs between 2.5 and 30 °C and at pH 6–10. The optimum temperature and pH for growth are 25 °C and 7.0. NaCl is tolerated at 5.8% and growth also occurs in the absence of salt. Positive in tests for catalase, oxidase, β-galactosidase, phosphatase, arginine dihydrolase, lysine decarboxylase, ornithine decarboxylase, arginine decarboxylase, gelatinase, caseinase, malonate utilization, aesculin hydrolysis and starch hydrolysis and in the Voges–Proskauer test. Negative for urease, lipase, DNase, tryptophan deamination, nitrate reduction to nitrite, citrate utilization, indole production, H₂S production and in the methyl red test. The following are utilized: D-glucose, D-fructose, D-galactose, D-lactose, maltose, D-mannose, L-rhamnose, raffinose, trehalose, D-ribose, melibiose, melezitose, cellobiose, sucrose, D-xylose, L-xylose, L-arabinose, D-arabinose, glycerol, D-sorbitol, dulcitol, starch, inulin, dextrin, myo-inositol, glycogen, sodium thioglycolate, methyl α-D-mannoside, amygdalin, arbutin, lactic acid, pyruvate, erythritol, γ-glucuronic acid, α-ketoglutaric acid, 5-ketogluconate, L-glycine, L-ornithine, L-alanine, L-leucine, L-isoleucine, L-lysine, L-arginine, L-glutamic acid, L-glutamine, L-asparagine, L-proline, L-histidine, L-creatinine. The following are not utilized: L-fucose, L-sorbose, D-mannitol, D-adonitol, hydroxybutyric acid, dextran, sodium gluconate, sodium fumarate, sodium formate, sodium succinate, sodium propionate, sodium acetate, polyethylene glycol, potassium acetate, cellulose, methyl α-D-glucoside, methyl-α-D-galactoside, methyl β-D-galactoside, N-acetylglucosamine, salicin, xylitol, citric acid, fumaric acid, L-malic acid, valeric acid, L-aspartic acid, L-cysteine, L-methionine, L-serine, L-threonine, L-tyrosine, L-tryptophan, L-valine and L-phenylalanine. Acid is produced from cellobiose, D-fructose,

D-glucose, D-mannose, maltose, D-ribose, L-rhamnose, sucrose, trehalose, L-xylose and inulin, but not from D-sorbitol, melibiose, D-lactose, L-arabinose or D-adonitol. Resistant to the following antibiotics (µg): amoxicillin (30), colistin (10), doxycycline (25), norfloxacin (10), nitrofurantoin (300) and sulfamethoxazole (50). Sensitive to the following antibiotics (µg): co-trimoxazole (25), clindamycin (25), amikacin (30), nalidixic acid (30), tobramycin (15), lomefloxacin (30), roxithromycin (30), ciprofloxacin (30), lincomycin (15), cefotaxime (30), cefazolin (30), kanamycin (30), novobiocin (30), chloramphenicol (30), ampicillin (25), tetracycline (30), streptomycin (25), erythromycin (15), bacitracin (10), gentamicin G (30), polymyxin B (50), oleandomycin (15), spectinomycin (100), rifampicin (25) and carbenicillin (100). The respiratory quinones are MK-6, MK-7 and MK-8 and the cell-wall peptidoglycan is of the Lys–Gly type. Phosphatidylglycerol, diphosphatidylglycerol, phosphatidylethanolamine, phosphatidylserine and phosphatidylinositol and an unidentified lipid are present. The fatty acids present are iso-C_{12:0} (2.0%), C_{12:0} (1.4%), iso-C_{13:0} (16.1%), anteiso-C_{13:0} (15.2%), iso-C_{14:0} (2.3%), C_{14:0} (1.3%), iso-C_{15:0} (17.1%), anteiso-C_{15:0} (6.2%), C_{15:1ω5c} (2.2%), iso-C_{16:0} (4.1%), C_{16:0} (3.6%), C_{16:1ω11c} (0.7%), anteiso-C_{17:1ω9c} (1.3%), iso-C_{17:0} (10.6%), iso-C_{17:1ω10c} (2.3%), anteiso-C_{17:0} (3.3%), C_{18:0} 10 methyl (1.1%), C_{19:0} (2.7%), summed feature 1 (1.8%), summed feature 3 (1.7%), summed feature 4 (0.8%) and summed feature 8 (2.2%).

The type strain, DVS 3Y^T (=MTCC 4816^T=JCM 14376^T), was isolated from a moraine sample from the McMurdo Dry Valleys, Antarctica. The DNA G+C content of the type strain is 48.5 mol%.

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References

- Chaturvedi, P. & Shivaji, S. (2006). *Exiguobacterium indicum* sp. nov., a psychrophilic bacterium from the Hamta glacier of the Himalayan mountain ranges of India. *Int J Syst Evol Microbiol* **56**, 2765–2770.
- Chaturvedi, P., Reddy, G. S. N. & Shivaji, S. (2005). *Dyadobacter hamtensis* sp. nov., from Hamta glacier, located in the Himalayas, India. *Int J Syst Evol Microbiol* **55**, 2113–2117.
- Collins, M. D., Pirouz, T., Goodfellow, M. & Minnikin, D. E. (1977). Distribution of menaquinones in actinomycetes and corynebacteria. *J Gen Microbiol* **100**, 221–230.
- Collins, M. D., Lund, B. M., Farrow, J. A. E. & Schleifer, K. (1983). Chemotaxonomic study of an alkalophilic bacterium, *Exiguobacterium aurantiacum* gen. nov., sp. nov. *J Gen Microbiol* **129**, 2037–2042.
- Farrow, J. A. E., Wallbanks, S. & Collins, M. D. (1994). Phylogenetic interrelationships of round-spore-forming bacilli containing cell walls based on lysine and the non-spore-forming genera *Caryophanon*, *Exiguobacterium*, *Kurthia* and *Planococcus*. *Int J Syst Bacteriol* **44**, 74–82.

- Felsenstein, J. (1993). PHYLIP (phylogeny inference package) version 3.5.1. Distributed by the author. Department of Genome Sciences, University of Washington, Seattle, USA.
- Fruhling, A., Schumann, P., Hippe, H., Straubler, B. & Stackebrandt, E. (2002). *Exiguobacterium undae* sp. nov. and *Exiguobacterium antarcticum* sp. nov. *Int J Syst Evol Microbiol* **52**, 1171–1176.
- Gee, J. M., Lund, B. M., Metcalf, G. & Peel, J. L. (1980). Properties of a new group of alkalophilic bacteria. *J Gen Microbiol* **117**, 9–17.
- Hugh, R. & Leifson, E. (1953). The taxonomic significance of fermentative versus oxidative metabolism of carbohydrates by various gram negative bacteria. *J Bacteriol* **66**, 24–26.
- Kim, I.-G., Lee, M.-H., Jung, S.-Y., Song, J. J., Oh, T.-K. & Yoon, J.-H. (2005). *Exiguobacterium aestuarii* sp. nov. and *Exiguobacterium marinum* sp. nov., isolated from a tidal flat of the Yellow Sea in Korea. *Int J Syst Evol Microbiol* **55**, 885–889.
- Kimura, M. (1980). A simple method for estimating evolutionary rates of base substitutions through comparative studies of nucleotide sequences. *J Mol Evol* **16**, 111–120.
- Komagata, K. & Suzuki, K. (1987). Lipid and cell-wall analysis in bacterial systematics. *Methods Microbiol* **19**, 161–207.
- Lányi, B. (1987). Classical and rapid identification methods for medically important bacteria. *Methods Microbiol* **19**, 1–67.
- Lopez-Cortes, A., Schumann, P., Pukall, R. & Stackebrandt, E. (2006). *Exiguobacterium mexicanum* sp. nov. and *Exiguobacterium artemiae* sp. nov. isolated from the brine shrimp *Artemia franciscana*. *Syst Appl Microbiol* **29**, 183–190.
- Reddy, G. S. N., Matsumoto, G. I. & Shivaji, S. (2003). *Sporosarcina macmurdoensis* sp. nov., from a cyanobacterial mat sample from a pond in the McMurdo Dry Valleys, Antarctica. *Int J Syst Evol Microbiol* **53**, 1363–1367.
- Rodrigues, D. F., Goris, J., Vishnivetskaya, T., Gilichinsky, D., Thomashow, M. F. & Tiedje, J. M. (2006). Characterization of *Exiguobacterium* isolates from the Siberian permafrost. Description of *Exiguobacterium sibiricum* sp. nov. *Extremophiles* **10**, 285–294.
- Sato, N. S. & Murata, N. (1988). Membrane lipids. In *Methods in Enzymology*, vol. 167, pp. 251–259, Edited by L. Packer & A. N. Glazer. New York: Academic Press.
- Shivaji, S., Rao, N. S., Saisree, L., Reddy, G. S. N., Seshu Kumar, G. & Bhargava, P. M. (1989). Isolates of *Arthrobacter* from the soils of Schirmacher Oasis, Antarctica. *Polar Biol* **10**, 225–229.
- Shivaji, S., Ray, M. K., Rao, N. S., Saisree, L., Jagannadham, M. V., Kumar, G. S., Reddy, G. S. N. & Bhargava, P. M. (1992). *Sphingobacterium antarcticus* sp. nov. a psychrotrophic bacterium from the soils of Schirmacher Oasis, Antarctica. *Int J Syst Bacteriol* **42**, 102–106.
- Shivaji, S., Vijaya Bhanu, N. & Aggarwal, R. K. (2000). Identification of *Yersinia pestis* as the causative organism of plague in India as determined by 16S rDNA sequencing and RAPD-based genomic fingerprinting. *FEMS Microbiol Lett* **189**, 247–252.
- Shivaji, S., Reddy, G. S. N., Raghavan, P. U. M., Sarita, N. B. & Delille, D. (2004). *Psychrobacter salsus* sp. nov. and *Psychrobacter adeliensis* sp. nov. isolated from fast ice from Adelie Land, Antarctica. *Syst Appl Microbiol* **27**, 628–635.
- Shivaji, S., Chaturvedi, P., Reddy, G. S. N. & Suresh, K. (2005). *Pedobacter himalayensis* sp. nov., from the Hamta glacier located in the Himalayan mountain ranges of India. *Int J Syst Evol Microbiol* **55**, 1083–1088.
- Smibert, R. M. & Krieg, N. R. (1994). Phenotypic characterization. In *Methods for General and Molecular Bacteriology*, pp. 607–654. Edited by P. Gerhardt. Washington, DC: American Society for Microbiology.
- Stackebrandt, E. & Goebel, B. M. (1994). Taxonomic note: a place for DNA-DNA reassociation and 16S rRNA sequence analysis in the present species definition in bacteriology. *Int J Syst Bacteriol* **44**, 846–849.
- Thompson, J. D., Higgins, D. G. & Gibson, T. J. (1994). CLUSTAL W: improving the sensitivity of progressive multiple sequence alignment through sequence weighting, position-specific gap penalties and weight matrix choice. *Nucleic Acids Res* **22**, 4673–4680.
- Tourova, T. P. & Antonov, A. S. (1987). Identification of microorganisms by rapid DNA-DNA hybridization. *Methods Microbiol* **19**, 333–355.
- Yumoto, I., Narisawa, M. H., Hirota, K., Shingyo, T., Takebe, F., Nodasaka, Y., Matsuyama, H. & Hara, I. (2004). *Exiguobacterium oxidotolerans* sp. nov., a novel alkaliphile exhibiting high catalase activity. *Int J Syst Evol Microbiol* **54**, 2013–2017.