NOTE

***Psychrobacter submarinus* sp. nov. and *Psychrobacter marincola* sp. nov., psychrophilic halophiles from marine environments**Lyudmila A. Romanenko,¹ Peter Schumann,² Manfred Rohde,³
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Two novel psychrophilic, halophilic, *Psychrobacter*-like bacteria, strains KMM 225^T and KMM 277^T, were isolated from sea water and the internal tissues of an ascidian *Polysyncraton* sp. specimen, respectively, and characterized using a polyphasic approach, which included phenotypic, genotypic, chemotaxonomic and phylogenetic analyses. The novel marine isolates were Gram-negative, aerobic, coccoid, oxidase- and catalase-positive, non-pigmented, non-motile, psychrophilic and halophilic and they utilized a restricted spectrum of carbon sources. Strains KMM 225^T and KMM 277^T required sea water or sodium ions for growth and were tolerant of up to 12–15% (w/v) NaCl. Growth of strains KMM 225^T and KMM 277^T was observed at 4–35 and 7–35 °C, respectively. The DNA G+C contents of KMM 225^T and KMM 277^T were respectively 46.8 and 50.7 mol%. Comparison of almost complete 16S rDNA sequences of strains KMM 225^T and KMM 277^T revealed that both strains were phylogenetically most closely related to each other (99.9% sequence similarity) and slightly less related to *Psychrobacter glacincola*, with 97.2 and 97.8% similarity, respectively. DNA–DNA reassociation between KMM 225^T and KMM 277^T revealed 15% similarity, whereas similarity to other *Psychrobacter* species was 14–25%. Strains KMM 225^T and KMM 277^T differed from one another in their growth temperature, organic substrate utilization, antibiotic sensitivity and DNA G+C content. Both strains examined could be distinguished from all previously described *Psychrobacter* species by their physiological, genotypic and phylogenetic characteristics. On the basis of the physiological and molecular properties of the novel isolates, the names *Psychrobacter submarinus* sp. nov. (type strain KMM 225^T = DSM 14161^T) and *Psychrobacter marincola* sp. nov. (type strain KMM 277^T = DSM 14160^T) are proposed.

Keywords: *Psychrobacter submarinus* sp. nov., *Psychrobacter marincola* sp. nov., psychrophilic, halophilic bacteria, marine environments

Gram-negative, aerobic, non-motile, psychrophilic, halotolerant micro-organisms are common inhabitants of terrestrial and marine environments, including the Antarctic and deep oceanic environments. The genus *Psychrobacter* was described to accommodate Gram-negative, moraxella-like, coccoid, oxidase-posi-

tive, non-motile, psychrophilic or psychrotolerant, halotolerant bacteria (Juni & Heym, 1986). DNA–rRNA hybridization analysis performed by Rossau *et al.* (1991) showed that this genus belongs to the family *Moraxellaceae*, which also includes the genera *Moraxella* and *Acinetobacter* (Rossau *et al.*, 1991). Members of the genus *Psychrobacter* have been isolated from a wide range of habitats including sea water, the gills and skin of fish, poultry, food and clinical sources (Juni, 1991). Recently, three Antarctic *Psychrobacter* species with low growth-temperature

The GenBank/EMBL/DBJ accession numbers for the 16S rDNA sequences of strains KMM 225^T and KMM 277^T are respectively AJ309940 and AJ309941.

optima were described, *Psychrobacter urativorans* and *Psychrobacter frigidicola*, both from Antarctic ornithogenic soils, and *Psychrobacter glacincola* from Antarctic sea ice (Bowman *et al.*, 1996, 1997), indicating that members of the genus *Psychrobacter* are widespread in cold Antarctic environments. The novel species *Psychrobacter pacificensis*, which differs from known terrestrial and Antarctic species, was described for psychrophilic strains isolated from deep sea water (5000–6000 m) of the Japan Trench (Maruyama *et al.*, 2000).

In the present study, the taxonomic position of two strains, KMM 225^T and KMM 227^T, from marine environments was defined. Results revealed that these bacteria represent two novel species of the genus *Psychrobacter*, for which the names *Psychrobacter submarinus* sp. nov. and *Psychrobacter marincola* sp. nov. are proposed.

Isolation, cultural characteristics and morphology

Strain KMM 225^T was isolated from a sea-water sample obtained at a depth of 300 m in the north-western part of the Pacific Ocean (12° 41' N, 132° 38' W) in July 1985. The sea-water sample was taken with a plastic hydrological bathometer. Strain KMM 277^T was isolated from the homogenized internal tissues of the ascidian *Polysyncrator* sp., collected in the Indian Ocean (33° 41' S, 134° 28' E) in March 1988. The homogenate was prepared and diluted in sterile sea water. Aliquots of the sea-water sample and the diluted homogenate were spread on agar plates of sea-water medium (SWM), which was made up of 750 ml sea water, 250 ml distilled water and the following components (g l⁻¹): peptone (5.0), yeast extract (2.5), glucose (1.0), K₂HPO₄ (0.2), MgSO₄ (0.05) and agar (15.0). Samples were incubated for 7–10 days at 28 °C. Isolates were cultivated aerobically on SWM, 2216 marine agar (MA) or marine broth (MB) (Difco) at 25–28 °C. Bacteria were stored at –80 °C in 30% (v/v) glycerol. Isolates designated KMM 225^T and KMM 277^T have been deposited in the Collection of Marine Micro-organisms (KMM) of the Pacific Institute of Bioorganic Chemistry, Vladivostok, Russia. For determination of biochemical and cultural tests, except those used for determination of the temperature range for growth, *Psychrobacter immobilis* CIP 102557^T, *P. urativorans* CIP 105100^T and *P. frigidicola* CIP 105101^T were cultivated on trypticase soy agar (TSA; Difco) at 28, 20 and 15 °C, respectively. *Psychrobacter phenylpyruvicus* CIP 82.27^T was grown on brain/heart infusion agar (BHIA; Difco) supplemented with 5% (v/v) blood serum at 30 °C and *P. glacincola* CIP 105313^T was grown on MA 2216 at 15 °C.

Cell morphology was examined by transmission electron microscopy of exponential-phase cells grown in MB 2216. Cells were fixed with 1% (v/v) glutaraldehyde and negatively stained with 4% (w/v) aqueous

Fig. 1. Transmission electron micrographs showing general morphology of negatively stained cells of *Psychrobacter submarinus* KMM 225^T (a, b) and *Psychrobacter marincola* KMM 277^T (c). Bars, 1 (a), 0.5 (b) and 0.25 (c) µm.

uranyl acetate and carbon film. Samples were examined with a Zeiss transmission electron microscope TEM910 at an acceleration voltage of 80 kV at calibrated magnifications. Motility was observed with a light microscope by the hanging-drop method.

The strains studied were slightly different in morphology and cell dimensions. Whereas cells of KMM 225^T were ovoid, 1.6–1.9 µm long and 0.7–1.0 µm in diameter (Fig. 1a, b), those of KMM 277^T were coccoid, 0.7–1.0 µm in diameter (Fig. 1c).

Growth characteristics and metabolic properties

The Gram-reaction, oxidase, catalase, production of caseinase, deoxyribonuclease, gelatinase and lipase (Tween 80) were tested according to the standard methods described by Smibert & Krieg (1994) using SWM as the basal medium. Amylase activity was observed on nutrient medium supplemented with starch at a concentration of 10% (w/v) by flooding plates with 1% (w/v) iodine solution. The ability to grow at different temperatures was tested on MA and TSA at 4, 7, 10, 15, 25, 30, 35, 37 and 40 °C. The pH range for growth (5.0–10.0) was tested using MB with pH values adjusted by the addition of 5 M NaOH or HCl. Sodium ion requirement and tolerance of NaCl were examined on SWM prepared with the artificial sea-water base adjusted so that the amount of NaCl was between 0 and 20% (w/v). Oxidative utilization of glucose was determined by the method of Leifson (1963). Leifson's medium was also used for testing acid production from sugars (with test sugars at 1%, w/v). Additional biochemical tests were performed using the API 20NE test kits (bioMérieux) as described by the manufacturer, except that strains were suspended in 3% (w/v) NaCl solution. All isolates were characterized physiologically by the Biolog GN MicroPlate method. Strains were grown for 24 h at 28 °C on medium MA 2216 and the microtitre plates were inoculated with cells suspended in 2.5% (w/v) NaCl. Results were read automatically with a spectrophotometer after 24 and 48 h incubation at 28 °C. Antibiotic sensitivity was tested by spreading bacterial suspensions on agar plates of TSA (for *P. immobilis* CIP 102557^T, *P. urativorans* CIP 105100^T and *P. frigidicola* CIP 105101^T), BHIA supplemented with 5% (v/v) blood serum (for *P. phenylpyruvicus* CIP 82.27^T) or MA (for *P. glacincola* CIP 105313^T, KMM 225^T and KMM 277^T) and applying discs impregnated with antibiotics (content per disc): ampicillin, 10 µg; benzylpenicillin, 10 U; gentamicin, 10 µg; kanamycin, 30 µg; carbenicillin, 25 µg; lincomycin, 15 µg; oleandomycin, 15 µg; polymyxin, 300 U; streptomycin, 30 µg; tetracycline, 30 µg; and neomycin, 15 µg.

The main phenotypic features of the novel isolates were characteristic of members of the genus *Psychrobacter*: Gram-negative, aerobic, oxidase- and catalase-positive, non-motile, non-pigmented, psychrophilic cocco-bacilli. On MA, colonies were whitish, circular with an entire margin, convex and 2–3 mm in diameter. Strains KMM 225^T and KMM 277^T grew at 4–35 and 7–35 °C, respectively, but not at 37–40 °C. The pH range for growth was 5.5–9.5, with optimum growth at pH 6.5–8.5. Both strains required sodium ions for growth and tolerated 12–15% (w/v) NaCl. The physiological and genomic characteristics of strains KMM 225^T and KMM 277^T were compared with those of other species of *Psychrobacter* (Table 1). In addition to the physiological characteristics that were negative for all *Psychrobacter* type strains (see Table 1), both strains were negative for urease, nitrate

reduction, tryptophan deaminase, phenylalanine deaminase, the ONPG test, Simmons' citrate and Voges–Proskauer tests, H₂S production, denitrification, acid production from D-glucose, D-xylose, rhamnose, galactose, maltose, lactose and mannitol and utilization of citrate, L-malate, lactate, acetate, propionate, L-histidine, L-proline, L-alanine and *n*-valerate.

Chemotaxonomy

Cellular fatty acid methyl esters obtained from cells grown in MB at 28 °C by the method of Stead *et al.* (1992) were separated by GC (Groth *et al.*, 1996) and identified as described previously (Schumann *et al.*, 1997). Polar lipids extracted by the method of Minnikin *et al.* (1979) were identified by two-dimensional TLC and spraying with specific reagents (Collins & Jones, 1980). The novel isolates possessed very similar whole-cell fatty acid profiles and the respective fatty acid contents (given in parentheses) of KMM 225^T and KMM 277^T were as follows: C_{10:0} (3.4 and 3.1%), C_{12:0} (7.1 and 4.4%), C_{16:1ω5c} (1.4 and 1.6%), C_{16:1ω7c} (4.1 and 3.4%), C_{18:1ω9c} (78.9 and 84.4%) and 3-OHC_{12:0} (2.1 and 3.1%). For strain KMM 225^T only, 3.2% C_{18:1ω7c} was detected in the cellular fatty acid composition. As with other *Psychrobacter* species (Bowman *et al.*, 1996, 1997; Maruyama *et al.*, 2000), C_{18:1ω9c} was the major fatty acid. Minor components are non-uniform in members of the genus, which makes it difficult to deduce a genus-characteristic pattern. The phospholipid composition of both strains was the same and included phosphatidylethanolamine, phosphatidylglycerol, diphosphatidylglycerol, phosphatidylinositol and phosphatidylserine.

Molecular analysis

DNA was isolated by the procedure of Marmur (1961). DNA base composition was determined as described by Marmur & Doty (1962) with modification as described by Owen *et al.* (1969). The DNA G+C contents of strains KMM 225^T and KMM 277^T were respectively 46.8 and 50.7 mol%. The value for strain KMM 277^T was higher than those of other *Psychrobacter* strains, which ranged between 41 and 47 mol%. A previous extensive genomic study of *P. immobilis* strains showed that some of them had G+C contents up to 49 mol% (Rossau *et al.*, 1991). DNA–DNA relatedness was measured spectrophotometrically using the hybridization method described by De Ley *et al.* (1970). The experiments, performed between strains KMM 225^T and KMM 277^T and four type strains of *Psychrobacter* species, revealed low DNA–DNA similarity values (14–25%; Table 2).

Genomic DNA extraction, PCR-mediated amplification of the 16S rDNA and sequencing of PCR products were carried out as described by Rainey *et al.* (1996). Purified PCR products were sequenced directly using the *Taq* DyeDeoxy Terminator Cycle

Table 1. Phenotypic characteristics of strains KMM 225^T and KMM 277^T and type strains of some *Psychrobacter* species

Taxa: 1, KMM 277^T; 2, KMM 225^T; 3, *P. immobilis*; 4, *P. urativorans*; 5, *P. frigidicola*; 6, *P. phenylpyruvicus*; 7, *P. glacincola*; 8, *P. pacificensis*. Data were taken from the present study or from Bowman *et al.* (1996) (*P. immobilis*, *P. urativorans*, *P. frigidicola*, *P. phenylpyruvicus*), Bowman *et al.* (1997) (*P. glacincola*) or Maruyama *et al.* (2000) (*P. pacificensis*). All species and strains were positive for: oxidase, catalase, growth at 7–15 °C and growth in 3.0–6.5 % (w/v) NaCl. All strains were negative for: growth at 40 °C, glucose fermentation, indole production, arginine dihydrolase, lysine decarboxylase, ornithine decarboxylase, hydrolysis of aesculin, starch, casein and gelatin and DNase. The following carbon and energy sources were not utilized by any strains: *N*-acetyl D-glucosamine, *m*-hydroxybenzoate, glycogen, phenylacetate, caprate, adipate, 2-ketogluconate, 5-ketogluconate, D-glucose, L-arabinose, D-mannitol, D-melibiose, L-fucose, L-rhamnose, ribose, salicin, sucrose, D-sorbitol, D-xylose, cellobiose, lactose, glycerol, *myo*-inositol, caprylate and malonate. Abbreviations for strains KMM 277^T and KMM 225^T are: +, positive; –, negative. Abbreviations for other species are: +, positive test reaction for 90–100 % strains; –, positive test reaction for 0–10 % strains; v+, test is positive for 11–89 % strains, type strain is positive; v–, test is positive for 11–89 % strains, type strain is negative; w, weak reaction; ND, not determined.

Property	1	2	3	4	5	6	7	8
Urease	–	–	v+	v+	–	+	v–	+
Nitrate reduction	–	–	v–	v–	–	–	v+	–
Tryptophan deaminase	–	–	v–	–	+	–	–	–
Phenylalanine deaminase	–	–	+	–	+	+	–	–
Hydrolysis of:								
Tween 40	–	+	+	–	–	–	+	ND
Tween 80	+	+	+	–	–	+	+	ND
Growth in NaCl:								
0	–	–	+	+	+	+	+	–
1 %	+	+	+	+	+	+	+	v–
8 %	+	+	+	+	+	+	+	–
10 %	+	+	v+	–	–	–	+	–
12 %	+	+	v+	–	–	–	v+	–
15 %	+	+	–	–	–	–	–	–
Growth at:								
4 °C	–	+	+	+	+	+	+	+
25 °C	+	+	+	+	–	+	–	+
30 °C	+	+	+	v+	–	+	–	+
35 °C	+	+	v+	–	–	+	–	+
37 °C	–	–	–	–	–	+	–	+
Acid production from:								
D-Glucose	–	–	+	–	–	–	–	+
D-Xylose	–	–	+	–	–	–	–	+
L-Arabinose	–	+	+	–	–	–	–	+
Fructose	–	+	–	–	–	–	–	–
Rhamnose	–	–	+	–	–	–	–	–
Galactose	–	–	+	–	–	–	–	–
Maltose	–	–	+	–	–	–	–	–
Utilization of:								
Trehalose	–	+	–	–	–	–	–	ND
Citrate	–	–	v–	–	–	+	v+	–
Acetate	–	–	+	v+	+	+	+	v–
L-Malate	–	–	+	+	+	+	–	+
Lactate	–	–	+	v+	–	+	v+	v–
Propionate	–	–	v+	–	–	+	+	–
L-Histidine	–	–	+	–	–	–	v+	+
L-Proline	–	–	+	+	+	+	+	+
L-Alanine	–	–	+	–	–	+	v+	v–
<i>n</i> -Valerate	–	–	+	v+	+	+	+	–
Sensitivity to antibiotics:*								
Ampicillin	+	+	–	w	+	+	–	ND
Benzylpenicillin	+	–	–	–	–	+	–	ND
Gentamicin	+	+	+	+	w	w	+	ND

Table 1 (cont.)

Property	1	2	3	4	5	6	7	8
Kanamycin	+	+	+	+	+	—	+	ND
Carbenicillin	+	+	—	+	+	w	—	ND
Lincomycin	+	—	—	—	—	—	—	ND
Oleandomycin	+	—	—	—	—	w	—	ND
Polymyxin	+	+	+	+	+	+	+	ND
Streptomycin	+	+	+	+	+	+	+	ND
Tetracycline	+	—	—	+	w	+	—	ND
Neomycin	+	+	+	+	+	w	+	ND
DNA G + C content (mol %)	50.7	46.8	44–47	44–46	41–42	43	43–44	43–44

* Data from present study.

Table 2. DNA–DNA similarity of strains KMM 225^T and KMM 277^T and some type strains of *Psychrobacter*

Strain	DNA G + C content (mol %)	DNA–DNA similarity (%):	
		KMM 225 ^T	<i>P. immobilis</i> CIP 102557 ^T
KMM 225 ^T	46.8	100	25
KMM 277 ^T	50.7	15	14
<i>P. immobilis</i> CIP 102557 ^T	45.7	25	100
<i>P. urativorans</i> CIP 105100 ^T	44.0	24	NT
<i>P. frigidicola</i> CIP 105101 ^T	41.7	20	NT
<i>P. glacincola</i> CIP 105313 ^T	44.7	22	22

NT, Not tested.

Sequencing kit (Applied Biosystems) according to the manufacturer's instructions. An Applied Biosystems 310 DNA genetic analyser was used for the electrophoresis of the sequence reaction products.

The almost complete 16S rDNA sequences of isolates KMM 225^T and KMM 277^T were aligned manually using the ae2 editor (Maidak *et al.*, 1997) with those of all currently available nucleotide sequences of representative *Psychrobacter* strains retrieved from GenBank and EMBL. The method of Jukes & Cantor (1969) was used to calculate evolutionary distances. Phylogenetic dendrograms were reconstructed according to the method of De Soete (1983) and the neighbour-joining and maximum-likelihood methods contained in the PHYLIP package (Felsenstein, 1993). The accession numbers of the reference strains used in the phylogenetic analysis are shown in Fig. 2. Sequence similarity of 16S rDNA determined for strains KMM 225^T and KMM 277^T was 99.9%. Lower values, ranging between 95.7 and 97.8%, were found for the 16S rDNA of the two isolates and the type strains of *Psychrobacter* species, which themselves range between 94.1 and 98.6% similarity. Different treeing algorithms (De Soete, 1983; Felsenstein, 1993) placed the novel strains on a separate subline of descent,

which is well separated from three other sublines containing *Psychrobacter* species (Fig. 2, displaying a neighbour-joining analysis). Except for the branching point of strains KMM 225^T and KMM 277^T, bootstrap values of branching points were generally below 80% significance and, hence, the question of the order in which the majority of *Psychrobacter* type strains are related to each other is not yet settled. The database includes two sequences of the type strain of *P. immobilis*, namely those of ATCC 43116^T and DSM 7229^T (96.7% similarity). None of the sequences is closely related to those of strains KMM 225^T and KMM 277^T, but the phylogenetic position of the type strain of *P. immobilis* needs careful re-examination.

Taxonomic conclusions

This study has shown that strains KMM 225^T and KMM 277^T can be assigned to the genus *Psychrobacter* according to the key features of the original description (Juni & Heym, 1986) and phylogenetic analyses. Phylogenetically, the two isolates differed from each other and from all known *Psychrobacter* species. 16S rDNA sequence analysis revealed that strains KMM 225^T and KMM 277^T were almost indistinguishable from each other (99.9% similarity) and closely related

Fig. 2. 16S rDNA dendrogram showing the position of strains KMM 225^T and KMM 277^T among *Psychrobacter* species. Bar, 10 inferred nucleotide substitutions per 100 nucleotides.

to *P. glacincola* (97.2 and 97.8% similarity, respectively) and other members of this genus (Fig. 2). However, DNA–DNA reassociation data indicated that they constitute separate species. A low similarity value of 15%, determined for strains KMM 225^T and KMM 277^T, as well as similarly low values of 14–25% determined for these two strains and type strains of other *Psychrobacter* species, indicate that strains KMM 225^T and KMM 277^T belong neither to the same species nor to any of the described species of *Psychrobacter*. All values are well below the value of 70% accepted for the distinction of different species (Wayne *et al.*, 1987). As shown in Table 1, there are sufficient differences between these two genospecies in their physiological properties to allow their phenotypic identification. The detailed compilation of metabolic and physiological properties in Table 1 also indicates that the two genospecies can be clearly distinguished from the validly described properties. On the basis of the physiological, genotypic and phylogenetic characteristics, it is concluded that isolates KMM 225^T and KMM 277^T represent two distinct species of the genus *Psychrobacter* for which the names *Psychrobacter submarinus* sp. nov. and *Psychrobacter marincola* sp. nov., respectively, are proposed.

Description of *Psychrobacter submarinus* sp. nov.

Psychrobacter submarinus (sub.ma.ri'nus. L. prep. sub under; L. adj. *marinus* of the sea; N.L. adj. *submarinus* from under the sea).

Aerobic, Gram-negative, non-motile, non-pigmented, non-spore-forming, ovoid cells, 1.6–1.9 µm long and 0.7–1.0 µm in diameter. Oxidase- and catalase-positive. Colonies on nutrient agar are whitish, circular with an entire margin, convex, 2–3 mm in diameter. No fluorescent pigments are formed. Moderately halophilic. Sea water is required for growth; growth occurs in the presence of 0.5–15.5% (w/v) NaCl. Psychrophilic. Temperature range for growth is 4–35 °C, with an optimum at 25–28 °C. Does not grow at 37 or 40 °C. pH range for growth is 5.5–9.5, with optimum growth at 6.5–8.5. Strain is negative for Simmons' citrate test, ONPG test, acetoin production, H₂S production and denitrification. Other physiological reactions are listed in Table 1. Susceptible to ampicillin (10 µg), gentamicin (10 µg), kanamycin (30 µg), carbenicillin (25 µg), polymyxin (300 U), streptomycin (30 µg) and neomycin (15 µg). Resistant to benzylpenicillin (10 U), lincomycin (15 µg), oleandomycin (15 µg) and tetracycline (30 µg). The major fatty acid is C_{18:1}ω9c. The phospholipids are phosphatidylethanolamine, phosphatidylglycerol, diphosphatidylglycerol, phosphatidylinositol and phosphatidylserine. The G + C content of the DNA is 46.7 mol% (determined by the thermal denaturation method). Strain KMM 225^T (= DSM 14161^T) is the type strain, isolated from sea water at 300 m deep in the Pacific Ocean.

Description of *Psychrobacter marincola* sp. nov.

Psychrobacter marincola (ma.ri.n.co'la. L. gen. n. *maris* of the sea; L. n. *incola* inhabitant; N.L. n. *marincola* inhabitant of the sea).

Aerobic, Gram-negative, non-motile, non-pigmented, non-spore-forming, coccoidal cells, 0.7–1.0 µm in diameter. Oxidase- and catalase-positive. Colonies on nutrient agar are whitish, circular with an entire edge, convex, 1–2 mm in diameter. No fluorescent pigments are formed. Moderately halophilic. Sea water is required for growth; can grow at 0.5–15.5% (w/v) NaCl. Psychrophilic. Growth occurs at 7–35 °C, with an optimum at 25–28 °C. Does not grow at 4, 37 or 40 °C. The pH range for growth is 5.5–9.5, with optimum growth at 6.5–8.5. Negative for Simmons' citrate test, Voges–Proskauer reaction, ONPG test, H₂S production and denitrification. Other physiological reactions are listed in Table 1. Susceptible to ampicillin (10 µg), benzylpenicillin (10 U), gentamicin (10 µg), kanamycin (30 µg), carbenicillin (25 µg), lincomycin (15 µg), oleandomycin (15 µg), polymyxin (300 U), streptomycin (30 µg), tetracycline (30 µg) and neomycin (15 µg). The major fatty acid is C_{18:1}ω9c. The phospholipids are phosphatidylethanolamine, phosphatidylglycerol, diphosphatidylglycerol, phosphatidylinositol and phosphatidylserine. The G + C content of the DNA is 50.7 mol% (determined by the thermal denaturation method). Strain KMM 277^T (= DSM 14160^T) is the type strain, isolated from internal tissues of the ascidian *Polysyncrator* sp., collected in the Indian Ocean.

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