Characterization of the marine propionate-degrading, sulfate-reducing bacterium *Desulfofaba fastidiosa* sp. nov. and reclassification of *Desulfomusa hansenii* as *Desulfofaba hansenii* comb. nov.

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A rod-shaped, slightly curved sulfate reducer, designated strain P2^T, was isolated from the sulfate–methane transition zone of a marine sediment. Cells were motile by means of a single polar flagellum. The strain reduced sulfate, thiosulfate and sulfite to sulfide and used propionate, lactate and 1-propanol as electron donors. Strain P2^T also grew by fermentation of lactate. Propionate was oxidized incompletely to acetate and CO₂. The DNA G + C content was 48.8 mol%. Sequence analysis of the small-subunit rDNA and the dissimilatory sulfite reductase gene revealed that strain P2^T was related to the genera *Desulfonema*, *Desulfococcus*, *Desulfosarcina*, ‘*Desulfobotulus*’, *Desulfofaba*, *Desulfomusa* and *Desulfofrigus*. These genera include incomplete as well as complete oxidizers of substrates. Strain P2^T shared important morphological and physiological traits with *Desulfofaba gelida* and *Desulfomusa hansenii*, including the ability to oxidize propionate incompletely to acetate. The 16S rRNA gene similarities of P2^T to *Desulfofaba gelida* and *Desulfomusa hansenii* were respectively 92.9 and 91.5 %. Combining phenotypic and genotypic traits, we propose strain P2^T to be a member of the genus *Desulfofaba*. The name *Desulfofaba fastidiosa* sp. nov. (type strain P2^T = DSM 15249^T = ATCC BAA-815^T) is proposed, reflecting the limited number of substrates consumed by the strain. In addition, the reclassification of *Desulfomusa hansenii* as a member of the genus *Desulfofaba*, *Desulfofaba hansenii* comb. nov., is proposed. A common line of descent and a number of shared phenotypic traits support this reclassification.

INTRODUCTION

Propionate is produced during fermentation of organic matter under anoxic conditions. In oxygen-free marine sediments, propionate is quantitatively the second-most important volatile fatty acid (Parkes *et al.*, 1993). Despite its importance, *in situ* concentrations of propionate are low, because it is efficiently consumed by sulfate-reducing bacteria. Until recently, all propionate-consuming sulfate reducers grouped in one phylogenetic lineage around the genus *Desulfobulbus* (Widdel & Pfennig, 1982; Isaksen &

Teske, 1996; Rabus *et al.*, 2000). Apart from a common phylogeny, the organisms also all oxidize propionate incompletely to acetate. However, within the past few years, two novel species of propionate-consuming sulfate reducers have been isolated that share with the ‘*Desulfobulbaceae*’ the incomplete oxidation of propionate but group phylogenetically with the completely oxidizing genera of the *Desulfonema/Desulfococcus/Desulfosarcina* assemblage. The two species were placed in different genera as *Desulfofaba gelida* and *Desulfomusa hansenii* (Knoblauch *et al.*, 1999; Finster *et al.*, 2001).

In this communication, we report on the isolation and characterization of a third isolate, designated strain P2^T, which shares a common phylogeny and physiology with both *Desulfofaba gelida* and *Desulfomusa hansenii*.

METHODS

Origin of the enrichment cultures. Sediment cores were collected in February 2001 in Aarhus Bay, a semi-enclosed embayment on the

Published online ahead of print on 28 November 2003 as DOI 10.1099/ijs.0.02820-0.

Abbreviation: DSR, dissimilatory sulfite reductase.

The GenBank/EMBL/DDBJ accession numbers for the 16S rDNA and partial DSR gene sequences of strain P2^T are AY268891 and AY268892.

Phase-contrast micrographs showing cells of strain P2^T and a DSR sequence-based phylogenetic tree are available as supplementary material in IJSEM Online.

east coast of Jutland, Denmark (56° 09' 20" N 10° 19' 24" E). The cores were taken at a water depth of 15 m. The salinity was 28.4‰ and the temperature was 6.3 °C. The sediment consisted of fine sand, silt and clay. Measurements of methane oxidation rates and sulfate reduction revealed that both rates peaked in the 170–200 cm depth interval. This zone was identified as the sulfate–methane transition zone. Sediment for enrichment cultures was withdrawn from that depth.

Enrichment and isolation. The enrichment medium was identical to DSMZ medium 193 with the following modifications. Firstly, sodium acetate solution was not added and, secondly, the vitamin solution used in the original medium was replaced by the following solutions: 1 ml vitamin solution (10 mg biotin, 3 mg folic acid, 15 mg pyridoxine hydrochloride dihydrate, 100 mg nicotinic acid, 50 mg calcium D-pantothenate, 40 mg *p*-aminobenzoic acid and 1.42 g Na₂HPO₄ dissolved in 1 l distilled water), 1 ml thiamin chloride solution (100 mg thiamin chloride and 3.56 g Na₂HPO₄ dissolved in 1 l distilled water) and 1 ml vitamin B₁₂ solution (50 mg cyanocobalamin dissolved in 1 l distilled water). In addition, 2 ml Na₂SeO₃·5H₂O solution (stock concentration 10 µM) and 2 ml Na₂WO₄ solution (stock concentration 10 µM) were added to 1 l medium. The medium was prepared as described in the DSMZ manual. The medium was distributed into 50-ml screw-capped bottles. CaCl₂ (10 mM final concentration), MgCl₂ (50 mM final concentration), propionate (15 mM final concentration) and sulfate (20 mM final concentration) were added from sterile stock solutions prior to inoculation. For the isolation of pure cultures, the modified deep agar dilution technique (Isaksen & Teske, 1996) was applied. The agar was washed three times in distilled water before use.

Physiological tests. Substrate utilization was studied in completely filled screw-capped bottles (50 ml) or Hungate tubes with a gas phase. Growth tests with CO and methane were performed in half-full 100-ml bottles sealed with butyl-rubber stoppers. Growth was determined microscopically and by following the production of hydrogen sulfide using the method of Cline (1969). Cultures showing growth were transferred to fresh medium containing the respective substrate at least three times to confirm the result. Cultures not showing growth after 1 month were maintained for at least 4 months and checked for growth on a regular basis. The presence of polyhydroxyalkanoates in the cells was investigated with the Nile blue stain (Ostle & Holt, 1982). Temperature experiments were carried out in duplicate in a temperature-controlled block at 11 different temperatures ranging from 8.8 to 37.4 °C. pH tolerance was tested in triplicate in media adjusted to 12 different pH values between 4.0 and 8.5. Growth rates were calculated from changes in OD₆₀₀. At pH values above 7.4, reliable OD measurements were no longer possible due to precipitation of medium components. Production of hydrogen sulfide was therefore measured in all tubes and used as an indicator of activity. Cell production was confirmed by microscopy with a non-growing control as reference. The salt requirement for growth was monitored at 11 different NaCl concentrations ranging from 6 to 38 g l⁻¹, eight different MgCl₂·6H₂O concentrations ranging from 5 to 68 mM and eight different CaCl₂·2H₂O concentrations ranging from 1 to 13 mM. Tests were carried out in duplicate. The effect of different salt concentrations was monitored by measuring H₂S production over time.

Chemical analyses. Sulfide was measured as described by Cline (1969). Sulfate was determined by suppressed ion chromatography according to Isaksen & Finster (1996). Propionate and acetate were analysed by ion-exclusion chromatography as described by Finster *et al.* (2001). The G+C content of genomic DNA was determined at the Identification Service of the DSMZ by Peter Schumann. DNA was isolated according to Visuvanathan *et al.* (1989) and purified as

described by Cashion *et al.* (1977) and the G+C content was determined by HPLC analysis (Mesbah *et al.*, 1989; Tamaoka & Komagata, 1984).

Microscopy. Samples for TEM were placed on a carbon-celloidin copper grid (200 mesh) and stained with a drop of 1% (w/v) uranyl acetate. Cells were observed with a JEOL 1200EX transmission electron microscope at 120 kV.

Nucleic acid extraction, PCR amplification and sequencing.

Nucleic acid was extracted using the FastDNA spin kit for various samples (Bio 101) according to the instructions of the manufacturer. The SSU rDNA was PCR-amplified and sequenced as described by Lane (1991). Almost complete 16S rDNA sequences were obtained with primers 26F/1390R (primer nomenclature refers to the 5'-ends of respective target sites on the 16S rDNA according to the *Escherichia coli* numbering of the 16S rRNA; Brosius *et al.*, 1981). PCR was performed at an annealing temperature of 57 °C with 25 cycles. PCR products were purified with the QIAquick PCR purification kit (Qiagen) according to the instructions of the manufacturer. Sequences were determined using the direct dideoxyribonucleotide chain-termination method with reverse transcriptase as described by Lane *et al.* (1985) using the kit from Amersham Pharmacia Biotech. The gel was run on an ALFexpress sequencer (Amersham Pharmacia Biotech) and sequences were handled with the ALFwin software (Amersham Pharmacia Biotech). A sequence of 1386 nucleotides was obtained after sequencing both strands with multiple primers.

The dissimilatory sulfite reductase (DSR) gene was amplified using primers designed by Wagner *et al.* (1998). Due to problems with non-specific annealing and low yields of the correct amplicon, the band of the correct length was cut out of an agarose gel and purified (QIAquick gel extraction kit; Qiagen) following the manufacturer's instructions. The DNA was ligated into a pCR-XL-TOPO vector and transformed into ONE SHOT *E. coli* cells according to the instructions of the manufacturer (TOPO-XL-pCR Cloning; Invitrogen); randomly selected clones were purified using a purification kit (QIAprep Spin Miniprep kit; Qiagen). Clones were amplified using a ThermoSequenase fluorescent cycle-sequencing kit (Amersham Pharmacia Biotech) and sequenced on an ALFexpress sequencer. A partial sequence of 876 nucleotides was obtained; 845 positions were used in the alignment.

Phylogenetic analysis. Sequence fragments were assembled manually in SEQPUP version 0.6 (Gilbert, 2002). Each resulting rDNA sequence was aligned with its closest relative using the on-line service of Ribosomal Database Project II (RDP-II) Sequence Aligner version 1.7 and compared to the GenBank and RDP-II databases using the BLAST algorithm (National Center for Biotechnology Information) and SEQUENCE MATCH version 2.7 (Maidak *et al.*, 2001; Wheeler *et al.*, 2002; Zhang *et al.*, 2000). Alignments were checked manually in SEQPUP. DSR gene sequences were aligned as nucleotide sequences or translated into an amino acid sequence before being aligned against their closest relatives. Phylogenetic analyses were performed with PAUP version 4.0b10. Only unambiguously aligned positions were used. The data matrix for the 16S rDNA sequences was analysed by distance-matrix (neighbour-joining), maximum-parsimony and maximum-likelihood approaches, whereas the DSR matrices were analysed by distance-matrix analysis only. Generally, bootstrap analysis was performed with 100 resamplings, except from the distance and maximum-parsimony algorithms analysing the relatedness of the SSU rRNA genes. Here, 1000 bootstraps were performed. Multifurcations were created at the appropriate basal node when branching patterns were only supported in less than 50% of the bootstrap resamplings.

RESULTS AND DISCUSSION

Enrichment and isolation

After 1 week, dividing cells and an increasing amount of sulfide were observed in a culture bottle containing propionate and sulfate. After several transfers, a stable mixed culture was obtained composed of three different, morphologically distinguishable cell types: large, curved rods, which were the dominant type, thin rods with endospores and coccoid cells. In deep agar dilutions, two types of colonies developed. One was white and fluffy and the other light brown, disk-shaped and smooth. Colonies were picked and transferred to fresh media. Prior to isolation, samples from the two colony types were withdrawn using Pasteur pipettes and tested for the presence of the DSR gene. The DSR gene is indicative of sulfate reduction potential. Only cells from the light-brown colonies were DSR positive. Cells producing white colonies were not studied further. After two additional series of deep agar dilution, several pure cultures of propionate-degrading sulfate reducers were obtained. All cultures were morphologically identical. They contained rod-shaped, slightly curved cells. One culture, designated strain P2^T, was chosen for detailed investigation.

Purity controls

Culture purity was checked microscopically prior to each transfer. In addition, purity of the isolate was tested in sulfate-free medium containing yeast extract (0.1 %, w/v), glucose (10 mM), fumarate (10 mM) and pyruvate (20 mM). There was no growth on this medium.

Cell morphology

Cells of strain P2^T were slightly curved, rod-shaped with round ends. They were 0.8–1.0 µm wide and 3–3.8 µm long (Fig. 1, Table 1). When grown with thiosulfate as electron acceptor, cells were longer than cells grown with sulfate as electron acceptor (Supplementary Figs A and B in IJSEM Online). Cells were motile by means of a single polar flagellum.

Phylogeny based on comparison of SSU rDNA

Strain P2^T is a member of the δ -subclass of the *Proteobacteria*. Phylogenetic analysis of 16S rDNA (Fig. 2) and the DSR gene sequence (Supplementary Fig. C) showed that strain P2^T is related to the *Desulfonema*/*Desulfococcus*/*Desulfosarcina* assemblage, which also includes the incompletely oxidizing genera '*Desulfobotulus*', *Desulfofaba*, *Desulfomusa* and *Desulforegula*. The overall branching pattern was supported by high bootstrap values in trees based on neighbour-joining, maximum-parsimony and maximum-likelihood algorithms. The closest relatives of strain P2^T were *Desulfofaba gelida* and *Desulfomusa hansenii*, with respective 16S rDNA sequence similarity of 92.9 and 91.5 %. The detailed branching pattern was, however, dependent on the algorithm. Using the maximum-likelihood algorithm,

Fig. 1. Transmission electron photomicrograph of strain P2^T grown on propionate with sulfate as electron acceptor and stained with 1 % (w/v) uranyl acetate. Bar, 1 µm.

strain P2^T formed a common line of descent with *Desulfofaba gelida* and *Desulfomusa hansenii*. The branching pattern was supported by a bootstrap value of 54 %. Interestingly, neither the neighbour-joining nor maximum-parsimony algorithm supported this branching pattern. In contrast, both algorithms supported a multifurcation as the best fit, indicating that a common ancestor of strain P2^T, *Desulfomusa hansenii* and *Desulfofaba gelida* could not be assigned.

Growth conditions and nutrition

Some properties of strain P2^T in comparison with other incomplete oxidizers of the *Desulfonema*/*Desulfococcus*/*Desulfosarcina* lineage are shown in Table 1. The optimal pH for growth was 6.8–7.1. The strain was able to grow in the pH range 6.0–7.75. The strain was grown at a salinity of 13.5 g l⁻¹; optimal growth was obtained in the range 10–16 g l⁻¹. However, the strain was able to grow at all salinities tested (6–38 g l⁻¹). CaCl₂ at a concentration of 7 mM enhanced growth and, again, growth was observed at all tested CaCl₂ concentrations (1–13 mM). MgCl₂ concentrations of 5–68 mM neither enhanced nor inhibited

Table 1. Physiological comparison of the incomplete oxidizers of the *Desulfonema/Desulfococcus/Desulfosarcina* assemblage

Strains: 1, '*Desulfobotulus sapovorans*' DSM 2055 (data from Postgate, 1984; Devereux *et al.*, 1989); 2, *Desulforegula conservatrix* DSM 13527^T (Rees & Patel, 2001); 3, *Desulfofrigus fragile* DSM 12345^T (Knoblauch *et al.*, 1999); 4, *Desulfofaba gelida* DSM 12344^T (Knoblauch *et al.*, 1999); 5, *Desulfomusa hansenii* DSM 12642^T (Finster *et al.*, 2001); 6, P2^T (this study). ND, No data; NA, not applicable; (+), slow growth. Butyrate, fructose, benzoate, alanine, 2-propanol, fumarate, valerate, butanol, glucose, nicotinate, acetate, ethanol, betaine, choline, pyruvate, succinate, sucrose, hydrogen, methylmercaptane, methane, acetate and CO were not utilized by strain P2^T. All taxa use sulfate (20 mM) and none of the taxa use nitrate (2 mM) as electron acceptors.

Characteristic	1	2	3	4	5	6
Cell shape	Vibrio	Rod	Curved rod	Curved thick rod	Curved thick rod	Curved thick rod
Width (µm)	1.5	1–1.3	0.8	3.1	2–3	0.8–1.7
Length (µm)	3.5–5.5	2.6–3	3.2–4.2	5.4–6.2	3–6	3.0–3.8
Motility	+	+	(+) (only old cultures)	(+) (only old cultures)	+	+
Temperature for growth (°C)						
Optimum	34	25–30	18	7	20	28
Range	15–38	ND–32	–1.8–ND	–1.8–10	8–30	5–33
Optimal pH	7.7	ND	7.0–7.4	7.1–7.6	7.2–7.4	6.8–7.1
Optimal salinity (%)	No optimum	1.0	1.0–2.5	1.4–2.5	1.3	1–1.6
Optimal MgCl ₂ (%)	ND	ND	0.3–2.0	0.015–2.2	1.0	No optimum*
Shortest doubling time (h) (on propionate)	NA	NA	NA	144	ND	4.3
Poly-hydroxyalkanoates	+	+	ND	ND	+	+
G + C content (mol%)	53	ND	52.1	52.5	53.4	48.8
16S rDNA similarity to strain P2 ^T (%)	89.3	91.6	91.8	92.9	91.5	NA
Propionate oxidation to acetate and CO ₂	–	–	–	+	+	+
Other electron donors	Fatty acids (≥C ₄), 2-methylbutyrate, lactate, pyruvate	Fatty acids (≥C ₄)	Fatty acids (≥C ₄), dicarboxylic acids, amino acids, alcohols	Butyrate, formate, lactate, pyruvate, other organic acids, alcohols, alanine	Pyruvate, alcohols, dicarboxylic acids, alanine, hydrogen + acetate	Lactate, 1-propanol
Fermentative growth	+	–	–	+	–	+
Fermentation of lactate	ND	–	–	–	–	+
Electron acceptors: (mM)						
Sulfite (5)	+	–	–	+	–	+
Thiosulfate (20)	–	–	–	+	–	+
Elemental sulfur (NA)	–	–	–	–	+	–

*Concentrations of 0.1–1.4 % were tested.

Fig. 2. Phylogenetic tree showing the affiliation of the 16S rDNA sequence from strain P2^T to selected reference sequences of the δ -subclass of the *Proteobacteria*. The tree was calculated by maximum-likelihood analysis with default settings of the distance-matrix analysis in PAUP 4.0b10. The sequences of *Synechococcus* sp. and *Nostoc muscorum* were used as an outgroup. Numbers at nodes indicate bootstrap values. Only bootstrap numbers greater than 50 are shown. Strains and GenBank accession numbers: *Desulfobacter latus* DSM 3381^T (AJ441315), *Desulfobacter postgatei* DSM 2034^T (AF418180), *Desulfobacter* sp. DSM 2035 (M34415), *Desulfobacterium autotrophicum* DSM 3382^T (M34409), '*Desulfobacterium vacuolatum*' DSM 3385 (M34408), '*Desulfobotulus sapovorans*' ATCC 33892 (M34402), *Desulfobulbus elongatus* FP^T (X95180), *Desulfobulbus rhabdoformis* M16^T (U12253), *Desulfococcus multivorans* DSM 2059^T (AF418173), *Desulfofaba gelida* PSv29^T (AF099063), *Desulfofrigus fragile* LSv21^T (AF099065), *Desulfofrigus oceanense* ASv26^T (AF099064), *Desulfomusa hansenii* DSM 12642^T (AF321820), *Desulfonema limicola* DSM 2076^T (U45990), *Desulforegula conservatrix* Mb1Pa^T (AF243334), strain oXyS1 (Y17286), *Desulfosarcina variabilis* DSM 2060^T (M34407), *Desulfovibrio desulfuricans* DSM 642^T (M37313), *Desulfovibrio profundus* DSM 11384^T (U90726), *Desulfovibrio zosteriae* DSM 11974^T (Y18049), *Nostoc muscorum* PCC 7120 (X59559), *Synechococcus* sp. PCC 6301 (X03538).

growth. The temperature range for growth was 5–33 °C. The isolate grew optimally at 28 °C with a doubling time of 4.3 h. With sulfate as electron acceptor, the isolate grew with propionate, lactate or 1-propanol as electron donors. Sulfate, sulfite and thiosulfate served as electron acceptors, while sulfur, nitrate, iron(III) citrate and fumarate did not. The strain also grew by fermentation of lactate. In contrast to *Desulfomusa hansenii* and *Desulfofaba gelida*, strain P2^T is not very versatile with respect to its range of substrates

(Table 1). Strain P2^T did not grow on pyruvate, succinate, fumarate, ethanol, butanol or alanine, substrates upon which *Desulfomusa hansenii* and *Desulfofaba gelida* grow. However, strain P2^T and *Desulfofaba gelida* but not *Desulfomusa hansenii* grow with thiosulfate as electron acceptor. In the assemblage of its closest relatives ('*Desulfobotulus*', *Desulfofrigus*, *Desulfomusa* and *Desulfofaba*), strain P2^T is distinct in its ability to ferment lactate, but fermentation in general is known from several members of the assemblage

(Widdel & Pfennig, 1984; Devereux *et al.*, 1989; Knoblauch *et al.*, 1999).

The three strains *Desulfomusa hansenii* DSM 12642^T, *Desulfofaba gelida* DSM 12344^T and strain P2^T were isolated from different habitats: *Desulfomusa hansenii* is a mesophilic bacterium isolated from the interior of surface-sterilized eelgrass roots, *Desulfofaba gelida* is a psychrophilic bacterium isolated from polar surface sediment and strain P2^T is a mesophilic bacterium isolated from the methane-sulfate transition zone 1.5 m below the sediment surface. Thus, we have evidence that anaerobic propionate degradation in sulfate-containing habitats is accomplished by at least two phylogenetically distinct lines of descent. Future studies should focus on the quantitative importance of the two lines in nature.

Storage compounds

Strain P2^T accumulated polyhydroxyalkanoates; cells of strain P2^T and *Desulfomusa hansenii* contained inclusions of polyhydroxyalkanoates of an unidentified type (K. Finster, unpublished data). *Desulfofaba gelida* was not tested for these compounds. The physiological role of these storage compounds has not been determined so far. The presence of polyhydroxyalkanoates, however, distinguishes them from *Desulfobulbus* species, which accumulate polyglucose instead (Stams *et al.*, 1983).

Stoichiometry of propionate oxidation

Changes in concentrations of propionate, acetate, sulfate and hydrogen sulfide and in the amount of cell carbon were measured in three different cultures. Propionate was oxidized incompletely to acetate and CO₂. One mol acetate was formed from 1 mol propionate and 1 mol hydrogen sulfide was formed from 1 mol sulfate. The molar ratios of propionate dissimilation to acetate and sulfide production agreed with the following equation:

The amount of propionate consumed for cell material synthesis was calculated by the equation given by Finster *et al.* (2001): $17\text{CH}_3\text{CH}_2\text{COO}^- + 5\text{HCO}_3^- + 15\text{H}_2\text{O} \rightarrow 14\text{C}_4\text{H}_7\text{O}_3 + 22\text{OH}^-$. Thus, 0.0118 mmol propionate is needed for production of 1.0 mg cells (dry weight). Strain P2^T produced 2.60 g biomass per mol propionate and thus incorporated about 3% of the consumed propionate into biomass. The inability of strain P2^T to oxidize acetate completely to CO₂ is interesting, considering its phylogenetic affiliation to the complete oxidizers such as *Desulfonema*, *Desulfococcus* and *Desulfosarcina*. Until recently, incompletely oxidizing sulfate reducers grouped separately from the complete oxidizers, with only one exception, '*Desulfobotulus sapovorans*' (= [*Desulfovibrio*] *sapovorans*) (Devereux *et al.*, 1989, 1990). In 1999, however, two incomplete sulfate reducers, *Desulfofaba gelida* and *Desulfofrigus*

fragile, whose closest relatives were members of the *Desulfosarcina* group, were isolated. In 2001, two additional incompletely oxidizing sulfate reducers, *Desulfomusa hansenii* and *Desulforegula conservatrix*, were described that were affiliated with the *Desulfosarcina* group (Finster *et al.*, 2001; Rees & Patel, 2001). The affiliation of incomplete oxidizers to the complete oxidizers is interesting, because the pathway used for degradation of the organic substrates differs markedly.

The completely oxidizing sulfate reducers use the classical β-oxidation pathway to degrade fatty acids (Janssen & Schink, 1995a, b). The oxidation of organic substrates by the incomplete oxidizer *Desulfobulbus propionicus* follows the reverse methylmalonyl-CoA pathway (Kremer & Hansen, 1988). Preliminary data indicate that *Desulfomusa hansenii* and strain P2^T use the same pathway as *Desulfobulbus propionicus*. Thus, despite the phylogenetic distance (Fig. 2), the pathway of propionate degradation seems to be shared among propionate-degrading sulfate reducers.

Taxonomic conclusions

We propose that strain P2^T (=DSM 15249^T=ATCC BAA-815^T) represents a novel species of the genus *Desulfofaba*, *Desulfofaba fastidiosa* sp. nov. In addition, based on its close phylogenetic affiliation to *Desulfofaba gelida* (Fig. 2) and the phenotypic similarities, we propose to reclassify *Desulfomusa hansenii* as *Desulfofaba hansenii* comb. nov. The description of the genus *Desulfofaba* must also be amended.

Emended description of the genus *Desulfofaba* Knoblauch *et al.* 1999

The original description of this genus was provided by Knoblauch *et al.* (1999). Additionally, some species are able to produce inclusions of polyhydroxyalkanoates. The type species is *Desulfofaba gelida*.

Description of *Desulfofaba hansenii* comb. nov.

Basonym: *Desulfomusa hansenii* Finster *et al.* 2001.

The description of this taxon was provided by Finster *et al.* (2001). The type strain is P1^T=DSM 12642^T=ATCC 700811^T.

Description of *Desulfofaba fastidiosa* sp. nov.

Desulfofaba fastidiosa (fas.ti.di.o'sa. L. fem. adj. *fastidiosa* fastidious, difficult to please, referring to the limited number of substrates used by the type strain).

Cells are curved, bean-shaped rods, 3–3.8 µm long and 0.8–1.0 µm wide when grown with sulfate, but longer when grown with thiosulfate. Cells are motile and have a single polar flagellum. Spores are not observed. Anaerobic growth with propionate, lactate or 1-propanol as electron donors and sulfate, sulfite or thiosulfate as electron acceptors. Growth is also possible by fermentation of lactate. The

pH range for growth is 6.0–7.75 (optimum 6.8–7.1). Temperature range for growth is 5–33 °C (optimum 28 °C). The DNA G+C content of the type strain is 48.8 mol%.

The type strain, P2^T (=DSM 15249^T=ATCC BAA-815^T), was isolated from the sulfate–methane transition zone (1.5 m below sediment surface) of Aarhus Bay (Denmark).

ACKNOWLEDGEMENTS

This study was supported by SNF grant no. 51-00-0309.

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