Desulfovibrio gabonensis sp. nov., a New Moderately Halophilic Sulfate-Reducing Bacterium Isolated from an Oil Pipeline

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Two moderately halophilic sulfate-reducing bacteria were isolated from an African oil pipeline and designated strains SEBR 3640 and SEBR 2840^T (T = type strain). Both of these strains possessed traits that define the genus *Desulfovibrio*. The cells of both isolates were motile curved rods that had a single polar flagellum and contained desulfovibrin, and both isolates utilized lactate, pyruvate, malate, fumarate, succinate, and ethanol in the presence of sulfate. Sulfite, thiosulfate, and elemental sulfur were also used as electron acceptors in the presence of lactate. However, both strains tolerated higher concentrations of NaCl (up to 17%) than all other *Desulfovibrio* species except *Desulfovibrio halophilus*, which tolerated a similar level of NaCl. The results of a 16S rRNA gene sequence analysis also placed the designated type strain, strain SEBR 2840, in the genus *Desulfovibrio* but revealed that this organism was significantly different from *D. halophilus* and all other validly described *Desulfovibrio* species. On the basis of our results, we propose that strain SEBR 2840^T is a member of a new species of the genus *Desulfovibrio*, *Desulfovibrio gabonensis*. The type strain of *D. gabonensis* is strain SEBR 2840 (= DSM 10636).

The presence of sulfate-reducing bacteria (SRB) in oil field waters has been recognized for a long time, and the activities of these microorganisms are closely linked to the history of oil. In 1926, Bastin et al. (3) studied the SRB communities in oil fields. After this pioneering work, several microbiological studies showed that SRB communities were widely distributed in most of the oil fields investigated (1, 5, 19).

In the course of a survey of different oil fields, we isolated several strains of SRB. Two strains, designated strains SEBR 2840^T (T = type strain) and SEBR 3640, were characterized further; these strains were isolated from water samples obtained from two oil pipelines transporting the oil produced at an offshore oil field to onshore facilities on the coast of Gabon (West Africa). Both strains were moderately halophilic, curved, rod-shaped organisms and on the basis of their morphological and physiological traits were assigned to the genus *Desulfovibrio*. The results of a 16S rRNA gene sequence analysis of strain SEBR 2840^T indicated that it was not related to any previously described *Desulfovibrio* species, and we propose that this strain should be assigned to a new species, *Desulfovibrio gabonensis*. Strain SEBR 2840 is the type strain of this species.

MATERIALS AND METHODS

Sources of the isolates. Strain SEBR 2840^T was isolated in February 1990 from a water sample taken from an oil pipeline linking offshore production platforms to onshore treatment facilities.

Strain SEBR 3640 was isolated 1 year later from another pipeline in the same offshore oil field.

Sampling and primary cultures of SRB were grown as described previously (4, 18). The total salinity of the culture media and the incubation temperature were

adjusted so that they corresponded to the in situ conditions (80 g/liter and 30°C, respectively).

Desulfovibrio halophilus and *Desulfohalobium retbaense* were provided by P. Caumette and B. Ollivier, respectively.

Media and culture conditions. SRB were isolated by the streak plate method, as described elsewhere (18). The culture medium used for isolation contained (per liter of distilled water) 2.0 g of Na₂SO₄, 0.3 g of KHCO₃, 1.0 g of CaCl₂ · 2H₂O, 60 g of NaCl, 6.0 g of MgSO₄ · 7H₂O, 1 ml of a 0.4% FeSO₄ · 7H₂O solution, 3.0 g of MOPS (morpholinopropane-sulfonic acid) buffer, 1.0 g of NH₄Cl, 6 ml of a 60% sodium lactate solution, 1.0 g of sodium acetate, 1.0 g of yeast extract, 0.5 g of cysteine hydrochloride, and 16 g of agar. The pH was adjusted to 7.4, and the medium was anaerobically distributed into bottles and autoclaved for 30 min at 105°C.

The final medium used for growth and maintenance of the strains was pre-

FIG. 1. Phase-contrast photomicrograph of *Desulfovibrio gabonensis* SEBR 2840^T grown with lactate as a carbon and energy source. Bar = 10 µm.

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FIG. 2. Transmission electron micrograph of *Desulfovibrio gabonensis* SEBR 2840^T after negative staining. Bar = 0.3 μ m.

pared as described by Pfennig et al. (24). It was supplemented with 50 g of NaCl per liter, 3.3 g of $\text{MgCl}_2 \cdot 6\text{H}_2\text{O}$ per liter, and 1.6 g of $\text{MgSO}_4 \cdot 7\text{H}_2\text{O}$ per liter.

The purity of strains was checked both by phase-contrast microscopy and by performing growth tests (both aerobically and anaerobically) in sulfate-free TYG medium (8).

Desulfovibrio halophilus and *Desulfohalobium retbaense* were grown as previously described (9, 21).

Morphology. An Olympus model BH-2 phase-contrast microscope was used for microscopy. Photomicrographs were obtained by using the agar slide methods (23). Cells were negatively stained by using 1% phosphotungstic acid, and electron photomicrographs were obtained with a JEOL model 1200 EX transmission electron microscope.

Metabolism and physiology. The physiological and metabolic tests used to characterize the strains included tests to determine utilization of different carbon substrates and energy sources, utilization of a variety of electron acceptors, fermentative growth, temperature, pH, and salinity optima, and vitamin requirements; the tests used have been described previously (18).

Pigments. The presence of desulfoviridin in cell extracts was determined by the method of Widdel and Pfennig (32). Briefly, cell extracts were prepared by ultrasonically disrupting bacterial cells for 15 min at 10 W with a Vibra Cell

72446 apparatus (Bioblock Scientific, Strasbourg, France) and then centrifuging the preparations at $25,000 \times g$ for 15 min and at $140,000 \times g$ for 90 min. The resulting cell extract was scanned with a UV spectrophotometer (Safas, Monaco, France).

Lipid analysis. Lipid extraction, esterification, fatty acid purification, and quantification by gas chromatography were performed as previously described (30). Briefly, lipids from lyophilized cells (50 to 100 mg) were extracted by using a modified Bligh-Dyer method (6, 29). The extracted lipids were fractionated into neutral lipids, glycolipids, and polar lipids by silicic acid column chromatography by using appropriate volumes of chloroform, acetone, and methanol, respectively. The phospholipids were subjected to mild alkaline methanolysis, and the resulting fatty acid methyl esters were purified by thin-layer chromatography and analyzed by gas chromatography and gas chromatography-mass spectrometry (15). The position and geometry of the double bond of each mono-saturated fatty acid were determined by using the dimethyl disulfide derivatives and the procedure described previously (15, 20).

Fatty acid nomenclature. In this paper a shorthand nomenclature is used, which is in the form of numbers separated by a colon. The number before the colon indicates the carbon chain length, and the number after the colon corresponds to the number of double bonds. The position of the double bond is

TABLE 1. Electron donors and acceptors utilized by *Desulfovibrio gabonensis* SEBR 2840^T and SEBR 3640

Electron donor or acceptor	Utilization by:	
	SEBR 2840 ^T	SEBR 3640
Electron donors^a		
H ₂ + CO ₂ (10 ⁵ Pa)	— ^b	—
H ₂ (10 ⁵ Pa) + acetate (10 mM)	+	+
Lactate (10 mM)	+	+
Pyruvate (10 mM)	+	+
Malate (10 mM)	+	+
Fumarate (10 mM)	+	+
Succinate (10 mM)	+	+
Formate (10 mM)	+	+
Acetate (10 mM)	—	—
Ethanol (10 mM)	+	+
Butanol (10 mM)	(+)	(+)
Electron acceptors^c		
Sulfate (20 mM)	+	+
Sulfite (10 mM)	+	+
Thiosulfate (10 mM)	+	+
Sulfur	+	+
Nitrate (10 mM)	—	—

^a Electron donors were tested in the presence of 30 mM sulfate. The following substrates were tested in the presence of sulfate but were not utilized: 10 mM glycerol, 10 mM butyrate, 5 mM isobutyrate, 5 mM valerate, 2 mM crotonate, 2 mM octanoate, 2 mM nonanoate, 5 mM palmitate, 2 mM tartrate, 5 mM benzoate, 5 mM citrate, 5 mM alanine, 10 mM lysine, 10 mM methionine, 5 mM cysteine, 5 mM glutamate, 5 mM aspartate, 5 mM betaine, 5 mM glycine, 0.25 M indole, 0.5 mM phenol, 2 mM nicotinate, 0.5 g of peptone per liter, 0.5 g of Casamino Acids per liter, and 0.5 g of yeast extract per liter.

^b —, no growth; +, good growth; (+), slight growth.

^c Electron acceptors were tested in the presence of 10 mM lactate as a carbon and energy source.

indicated by the symbol ω, followed by the number of carbons from the methyl end. The geometry of the double bonds is indicated by *cis* or *trans*.

G+C content of the DNA. The G+C content of strain SEBR 2840^T DNA was determined by high-performance liquid chromatography at the Identification Service of the Deutsche Sammlung von Mikroorganismen und Zellkulturen, Braunschweig, Germany.

16S rRNA sequencing. The 16S rRNA gene of SEBR 2840^T was amplified by adding 1 µl of cell culture to a thermocycler microtube containing 5 µl of 10× *Taq* buffer (Pharmacia, Uppsala, Sweden), 100 ng of each primer, and 3 µl of 25 mM MgCl₂ · 6H₂O. The volume was adjusted to 47.5 µl with sterile distilled water.

The two primers which we used (5'-AGAGTTTGATCCTGGCTCAG-3' and 5'-AAGGAGGTGATCCAGCC-3') were described by Weisburg et al. (28). The samples were denatured by heating them for 30 min at 92°C, and a mixture containing 2 µl of deoxynucleoside triphosphates (each at a concentration of 5 mM) and 0.5 µl of *Taq* polymerase (Pharmacia) was added. The samples were then placed in a Hybrid thermal reactor thermocycler and subjected to 35 cycles, each of which consisted of 1 min at 93°C, 1 min at 53°C, and 2.5 min at 72°C; this was followed by a final elongation step consisting of 10 min at 72°C.

The PCR products were purified by using the spun-column chromatography procedure and a Sephadryl S400 gel (Pharmacia). The purified products were sequenced directly with an Applied Biosystems model 373 automated DNA sequencer by using a prism dideoxy terminator cycle sequencing kit (Applied Biosystems, Ltd., Foster City, Calif.). The primers used were the primers used for PCR amplification and internal primers derived from partial sequence data.

The 16S rRNA genes of *Desulfohalobium retbaense* and *Desulfovibrio halophilus* were amplified, purified, and sequenced as previously described (10, 17, 26).

A search for sequences similar to the 16S rRNA sequence of SEBR 2840^T was performed by using the SIMILARITY-RANK tool of the Ribosomal Databank Project (22), the blast (2) and fasta (33) software, and the GenBank and EMBL databases. Sequences were aligned manually with an alignment editor, ac2. Positions of sequence and alignment uncertainty were omitted from the analysis, and pairwise evolutionary distances were determined by using the correction factor of Jukes and Cantor (16). Dendrograms were constructed from evolutionary distances by using the neighbor-joining package that forms part of the

PHYLP package of software programs (14). The tree topology was reexamined by using 100 bootstrapped data sets.

Nucleotide sequence accession numbers. The sequences of the 16S rRNA genes of strain SEBR 2840^T, *Desulfohalobium retbaense*, and *Desulfovibrio halophilus* have been deposited in the GenBank and EMBL databases under accession numbers U31080, U48244, and U48243, respectively.

RESULTS

Cell morphology. The cells of strains SEBR 2840^T and SEBR 3640 were actively motile vibrioid cells that occurred singly or in pairs (Fig. 1). Each cell moved by means of a polar flagellum (Fig. 2). Sporulation was never observed. The cells were 0.4 µm wide and 2 to 4 µm long. Gram staining was negative.

Physiological properties. The substrates tested as possible energy and carbon sources in the presence of sulfate as an electron acceptor are listed in Table 1. Strains SEBR 2840^T and SEBR 3640 grew well on lactate, pyruvate, malate, fumarate, succinate, and ethanol. Lactate was incompletely oxidized to acetate and CO₂. Slight growth occurred in the presence of butanol, and small amounts of sulfide were produced. H₂ and formate were used as electron donors in the presence of acetate. CO₂ could not be used as a sole source of carbon by either strain. The ability to use different electron acceptors was tested in minimal medium supplemented with lactate. Both strains reduced sulfate, sulfite, thiosulfate, and sulfur to sulfide. Nitrate could not be used.

In sulfate-free medium, strains SEBR 2840^T and SEBR 3640 grew well with malate, fumarate, and pyruvate as sole carbon and energy sources. Glucose, fructose, succinate, lactate, cysteine, and glycerol were never fermented.

In synthetic medium that was supplemented with lactate and sulfate and contained only 0.01% yeast extract, both strains grew in the presence of 1 to 17% NaCl. Optimum growth of strain SEBR 2840^T occurred in the presence of 5 to 6% NaCl, and optimum growth of strain SEBR 3640 occurred in the presence of 6 to 8% NaCl. The optimum pH was 6.9 to 7.3, and strain SEBR 2840^T grew well at pH values between 6.4 and 8.2. The optimum temperatures were 30°C for strain SEBR 2840^T and 38°C for strain SEBR 3640. These strains were mesophilic organisms; no growth occurred at 50°C or above, and both strains grew well at 20°C. Both strains did not grow without

TABLE 2. Fatty acid profiles of *Desulfovibrio gabonensis* SEBR 2840^T and SEBR 3640

Fatty acid	% in:	
	SEBR 2840 ^T	SEBR 3640
iso C _{14:0}	5.21	5.62
C _{14:0}	14.63	12.85
iso C _{15:1ω7}	0.30	0.33
iso C _{15:0}	7.90	9.22
anteiso C _{15:0}	38.44	42.45
C _{15:0}	1.01	1.36
iso C _{16:0}	0.78	0.87
C _{16:1ω9}		0.77
C _{16:1ω7c}	0.84	0.22
C _{16:0}	8.85	6.74
iso C _{17:0}	0.08	0.08
anteiso C _{17:0}	0.20	0.25
C _{17:0}	0.52	0.71
iso C _{18:0}	0.26	0.25
C _{18:1ω7c}	5.91	4.89
C _{18:1ω7t}		0.25
C _{18:1ω5c}		0.38
C _{18:0}	15.06	12.77

TABLE 3. Evolutionary similarity matrix

Species	% Sequence similarity																						
	<i>Agrobacterium tumefaciens</i>	<i>Escherichia coli</i>	<i>Pseudomonas testosteroni</i>	<i>Bdellovibrio stolpii</i>	<i>Myxococcus xanthus</i>	<i>Geobacter metallireducens</i>	<i>Desulfuromonas acetoxidans</i>	<i>Desulfomonile tiedjei</i>	<i>Desulfosarcina variabilis</i>	<i>Desulfobacter postgatei</i>	<i>Desulfomicrobium baculatum</i>	<i>Desulfomicrobium escambium</i>	<i>Desulfovibrio salexigens</i>	<i>Desulfovibrio longus</i>	<i>Desulfovibrio africanus</i>	<i>Desulfovibrio gigas</i>	<i>Desulfovibrio desulfuricans</i>	<i>Desulfovibrio piger</i>	<i>Desulfovibrio vulgaris</i>	<i>Desulfovibrio longreacheensis</i>	<i>Desulfovibrio gabonensis</i>	<i>Desulfovibrio halophilus</i>	
<i>Escherichia coli</i>	80.8																						
<i>Pseudomonas testosteroni</i>	78.4	81.2																					
<i>Bdellovibrio stolpii</i>	80.4	79.0	77.3																				
<i>Myxococcus xanthus</i>	81.1	79.5	77.5	81.4																			
<i>Geobacter metallireducens</i>	81.4	83.5	80.0	83.0	84.8																		
<i>Desulfuromonas acetoxidans</i>	82.3	81.6	79.2	83.9	84.5	90.6																	
<i>Desulfomonile tiedjei</i>	81.4	81.4	79.5	83.1	84.2	87.6	87.1																
<i>Desulfosarcina variabilis</i>	81.7	81.4	78.0	82.9	82.9	86.3	86.6	86.9															
<i>Desulfobacter postgatei</i>	79.0	79.5	77.7	82.6	81.0	84.9	85.3	83.6	87.8														
<i>Desulfomicrobium baculatum</i>	82.1	80.5	77.7	82.5	80.9	85.5	83.9	82.3	84.9	83.9													
<i>Desulfomicrobium escambium</i>	82.6	81.4	78.3	82.9	81.5	86.4	84.4	82.9	85.0	84.4	97.9												
<i>Desulfovibrio salexigens</i>	81.5	80.2	77.5	81.6	80.9	83.1	83.9	81.5	82.6	82.2	87.3	87.2											
<i>Desulfovibrio longus</i>	80.8	80.3	77.1	82.2	80.7	83.4	84.1	83.2	84.1	83.1	87.7	87.5	90.0										
<i>Desulfovibrio africanus</i>	79.8	81.1	78.7	81.1	82.1	84.3	84.3	81.4	83.9	83.1	88.2	88.5	88.4	88.6									
<i>Desulfovibrio gigas</i>	79.7	80.4	78.1	81.6	80.9	83.9	85.0	80.8	83.7	83.4	87.2	87.2	87.2	88.0	87.9								
<i>Desulfovibrio desulfuricans</i>	80.7	80.7	77.1	81.7	80.7	84.4	84.3	81.8	82.8	83.6	88.7	89.0	87.9	89.9	88.7	89.3							
<i>Desulfovibrio piger</i>	79.8	80.1	77.8	81.7	80.3	83.5	84.3	80.8	82.9	83.5	87.2	88.0	86.8	88.5	87.1	89.2	94.7						
<i>Desulfovibrio vulgaris</i>	80.1	80.2	77.6	79.8	78.8	84.3	83.5	80.3	83.5	81.2	87.0	87.3	89.0	88.9	89.4	87.3	91.2	90.7					
<i>Desulfovibrio longreacheensis</i>	80.9	80.3	77.4	81.0	80.3	84.3	83.9	80.5	83.5	82.3	87.5	88.2	88.2	88.9	88.3	89.6	92.1	92.1	94.5				
<i>Desulfovibrio gabonensis</i>	79.2	79.6	77.6	81.1	81.1	83.6	83.1	80.9	83.4	81.8	86.6	87.6	90.0	89.0	89.2	91.1	88.6	88.2	89.3	90.3			
<i>Desulfovibrio halophilus</i>	79.5	79.8	76.8	80.5	79.4	81.7	82.2	81.5	82.8	80.7	86.2	86.2	88.5	88.7	86.9	85.1	86.4	85.0	86.5	86.5	88.7		
<i>Desulfobalobium retbaense</i>	81.1	80.5	77.8	79.5	80.1	83.9	83.1	81.4	82.4	81.7	86.6	87.2	86.3	84.6	84.6	86.9	85.5	85.2	84.6	85.1	87.1	84.1	

vitamins (V7 solution [24]) or yeast extract in the culture medium after five transfers. Under optimum growth conditions, the doubling time of each culture was 10 h.

Pigment. Spectra of the soluble fraction exhibited the characteristic absorption band of desulfovibrin at 631 nm.

Lipid characteristics. The fatty acid profiles are shown in Table 2. Branched-chain saturated fatty acids accounted for 53 to 58% of the total fatty acids, with anteiso $C_{15:0}$ predominating (this fatty acid accounted for up to 40% of the total fatty acids). With the exception of $C_{18:1\omega7c}$ (which accounted for 5% of the total fatty acids), only traces of monounsaturated fatty acids were detected in both isolates.

G+C content. The G+C content of the DNA of strain SEBR 2840^T was 59.5 mol%.

16S rRNA gene sequence analysis. We sequenced 1,417 bases from positions 52 to 1461, 1,543 bases from positions 8 to 1542, and 1,541 bases from positions 9 to 1542 (*Escherichia coli* numbering of Winker and Woese [34]) of the 16S rRNA genes of strain SEBR 2840^T, *Desulfobalobium retbaense*, and *Desulfovibrio halophilus*, respectively. The results of our 16S rRNA gene sequence analysis indicated that strain SEBR 2840^T clustered with members of the genus *Desulfovibrio* (average level of similarity, 89%) and that its closest relative was *Desulfovibrio gigas* (level of similarity, 91%) (Table 3 and Fig. 3). Further analysis revealed that strain SEBR 2840^T was distantly related to the halophilic sulfate-reducing bacteria *Desulfovibrio halophilus* (level of similarity, 88.7%), *Desulfobalobium retbaense* (level of similarity, 87%), and *Desulfovibrio salexigens* (level of similarity, 90%).

DISCUSSION

Strains SEBR 2840^T and SEBR 3640, which were isolated from the production water of an oil field, are strictly anaerobic, slightly curved, rod-shaped organisms that use sulfate as an electron acceptor. The presence of desulfovibrin and the ability of these strains to use lactate and alcohols strongly suggest that they are affiliated with the genus *Desulfovibrio* in the

FIG. 3. Phylogenetic tree based on a comparison of the 16S ribosomal DNA sequences of *Desulfovibrio gabonensis* SEBR 2840^T and selected bacteria. α, β, γ, and δ are subdivisions of the Proteobacteria.

ecophysiological group containing the SRB (24). Because the two strains have very similar physiological and metabolic characteristics, they were considered members of the same species, and strain SEBR 2840 was chosen as the type strain. Both strains require NaCl for growth, like the four halophilic *Desulfovibrio* species described previously, *Desulfovibrio desulfuricans* subsp. *aestuarii* (25, 31), *Desulfovibrio giganteus* (13), *Desulfovibrio salexigens* (25, 31), and *Desulfovibrio halophilus* (9). Strains SEBR 2840^T and SEBR 3640 can easily be distinguished from the first three of these species by their NaCl tolerance. *Desulfovibrio desulfuricans* subsp. *aestuarii*, *Desulfovibrio giganteus*, and *Desulfovibrio salexigens* are slightly halophilic sulfate reducers (optimal growth occurs at salinities ranging from 1 to 4%); in contrast, both strain SEBR 2840^T and strain SEBR 3640 are moderately halophilic SRB that grow optimally in the presence of 5 to 8% NaCl and are able to grow in the presence of up to 17% NaCl. The ability to grow in the presence of such high NaCl concentrations has been observed previously with only two SRB, *Desulfovibrio halophilus* (9) and *Desulfohalobium retbaense* (21). *Desulfohalobium retbaense* is a motile, straight, rod-shaped organism that grows optimally in the presence of 10% NaCl, incompletely oxidizes lactate, ethanol, and pyruvate, and contains desulforubidin as a bisulfite reductase. Thus, morphologically and physiologically, *Desulfohalobium retbaense* is different from *Desulfovibrio* sp. strains SEBR 2840^T and SEBR 3640. Both strain SEBR 2840^T and strain SEBR 3640 are more closely related to *Desulfovibrio halophilus* than to other halophilic SRB. However, they differ from *Desulfovibrio halophilus* by their ability to oxidize fumarate, succinate, and malate; fermentative growth with malate, fumarate, and pyruvate has not been observed in *Desulfovibrio halophilus* (Table 4).

Moreover, the phylogenetic data showed that the three halophilic SRB that have been isolated so far are not closely related. *Desulfovibrio halophilus* and strain SEBR 2840^T are affiliated with the *Desulfovibrionaceae* (11) but are members of two different lines. Strain SEBR 2840^T is closely related to *Desulfovibrio gigas*, whereas *Desulfovibrio halophilus* belongs to a cluster that includes *Desulfovibrio longus* and *Desulfovibrio salexigens*.

The fatty acid profiles of isolates SEBR 2840^T and SEBR 3640 are similar in that branched-chain saturated fatty acids predominate and anteiso C_{15:0} is a major fatty acid. iso C_{15:0}, anteiso C_{15:0}, and C_{17:0} are found in many bacteria, including some aerobic and anaerobic bacteria, like the sulfate reducers (7, 12). iso C_{15:0} is the dominant fatty acid in most *Desulfovibrio* species and in the genera *Desulfomicrobium* and *Desulfomonas*. The percentage of this fatty acid is significantly lower in some other SRB (*Desulfovibrio gigas*, *Desulfovibrio giganteus*, and *Desulfovibrio sulfodismutans*), which have anteiso C_{15:0} as their major fatty acid. iso C_{17:1ω7}, which is not present in isolates SEBR 2840^T and SEBR 3640, is usually recognized as a biomarker for the genus *Desulfovibrio*, although *Desulfovibrio gigas* DSM 1382 does not contain this monounsaturated acid (27).

The metabolic and physiological characteristics of *Desulfovibrio* sp. strains SEBR 2840^T and SEBR 3640 are different enough from the characteristics of all previously described *Desulfovibrio* species that these two strains could be considered members of a new species.

This possibility is supported by the 16S rRNA gene sequence data, which clearly showed that strain SEBR 2840^T belongs to a cluster composed of all of the *Desulfovibrio* species examined so far. The evolutionary distances between strain SEBR 2840^T and other members of the genus *Desulfovibrio* are large enough to consider strain SEBR 2840^T a member of a new species of

TABLE 4. Major characteristics of moderately halophilic *Desulfovibrio* species

Characteristic	<i>Desulfovibrio gabonensis</i> SEBR 2840 ^T	<i>Desulfovibrio halophilus</i> SL 8903 ^a
Cell size (μm)	0.4 × 2-4	0.6 × 2.5-5
Salinity range (%)	1-17	3-18
Optimum salinity (%)	5-6	6-7
Desulfoviridin	+	+
G+C content (mol%)	59.5	60.7
Vitamins required	+	—
Optimum temp (°C)	30	35
Utilization of electron donors in the presence of sulfate		
H ₂ + CO ₂	— ^b	—
H ₂ + acetate	+	+
Lactate	+	+
Pyruvate	+	+
Malate	+	—
Fumarate	+	—
Succinate	+	—
Formate + acetate	+	+
Acetate	—	—
Ethanol	+	+
Isopropanol	—	—
Butanol	(+)	—
Fermentation in the absence of sulfate		
Pyruvate	+	—
Malate	+	—
Fumarate	+	—
Utilization of electron acceptors with lactate as an energy and carbon source		
Sulfate	+	+
Sulfite	+	+
Thiosulfate	+	+
Sulfur	+	(+)
Nitrate	—	—

^a Data from reference 9.

^b —, no growth; +, good growth; (+), slight growth.

this genus. Therefore, strains SEBR 2840^T and SEBR 3640 are considered representatives of a new species named *Desulfovibrio gabonensis*.

Description of *Desulfovibrio gabonensis* sp. nov. *Desulfovibrio gabonensis* (ga. bo' nen. sis. M. L. adj. *gabonensis*, pertaining to Gabon, a country in West Africa). Curved rod-shaped cells are 0.4 μm wide and 2 to 4 μm long. Gram negative. Motile by means of a polar flagellum. The pH range is 6.4 to 8.2; optimum growth occurs at pH 6.9 to 7.3. The temperature range is 15 to 40°C; optimum growth occurs at 30°C. The salinity range is 1 to 17%; optimum growth occurs in the presence of 5 to 6% NaCl.

Strictly anaerobic. Reduces sulfate, sulfite, thiosulfate, and elemental sulfur with production of sulfide. Nitrate is not used as an electron acceptor. The substrates that are oxidized by anaerobic respiration of sulfur compounds are H₂, formate, lactate, pyruvate, malate, fumarate, succinate, and ethanol. H₂ and formate are used in the presence of acetate. Slight growth occurs with butanol. Malate, fumarate, and pyruvate are fermented. Vitamins are required as growth factors. Desulfoviridin is present. The G+C content of the DNA is 59.5 mol%. The major lipids are anteiso C_{15:0} (40% of the total fatty acids), C_{18:0} (15%), C_{14:0} (14%), and C_{16:0} (8%). The 16S rRNA gene sequence has been deposited in the GenBank and EMBL databases under accession number U31080.

Habitat: production fluid from an oil-producing well in Gabon.

The type strain is strain SEBR 2840, which has been deposited in the Deutsche Sammlung von Mikroorganismen und Zellkulturen as strain DSM 10636.

ACKNOWLEDGMENTS

We are grateful to S. Jagoueix and P. Jara for helpful discussions about PCR. We thank B. Delpech, O. Bonin, and X. Dumont for helpful technical assistance during ribosomal DNA sequencing.

This research was supported by a grant from Elf Aquitaine (to C.T.-J.) and in part by a grant from the Australian Research Council (to B.K.C.P.).

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