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Geovibrio ferrireducens, a phylogenetically distinct dissimilatory Fe(III)-reducing bacterium

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Abstract A new, phylogenetically distinct, dissimilatory, Fe(III)-reducing bacterium was isolated from surface sediment of a hydrocarbon-contaminated ditch. The isolate, designated strain PAL-1, was an obligately anaerobic, nonfermentative, motile, gram-negative vibrio. PAL-1 grew in a defined medium with acetate as electron donor and ferric pyrophosphate, ferric oxyhydroxide, ferric citrate, Co(III)-EDTA, or elemental sulfur as sole electron acceptor. PAL-1 also used proline, hydrogen, lactate, propionate, succinate, fumarate, pyruvate, or yeast extract as electron donors for Fe(III) reduction. It is the first bacterium known to couple the oxidation of an amino acid to Fe(III) reduction. PAl-1 did not reduce oxygen, Mn(IV), U(VI), Cr(VI), nitrate, sulfate, sulfite, or thiosulfate with acetate as the electron donor. Cell suspensions of PAL-1 exhibited dithionite-reduced *minus* air-oxidized difference spectra that were characteristic of *c*-type cytochromes. Analysis of the 16S rRNA gene sequence of PAL-1 showed that the strain is not related to any of the described metal-reducing bacteria in the Proteobacteria and, together with *Flexistipes sinusarabici*, forms a separate line of descent within the Bacteria. Phenotypically and phylogenetically, strain PAl-1 differs from all other described bacteria, and represents the type strain of a new genus and species, *Geovibrio ferrireducens*.

Key words *Geovibrio* · Fe(III) reduction · Co(III) reduction · Sulfur reduction

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Introduction

Geological studies showing a strong correlation between isotopically light carbonates and magnetite accumulations in the Precambrian Banded Iron Formations have suggested that organic matter mineralization was coupled to Fe(III) reduction in the Precambrian biosphere (Walker 1984; Baur et al. 1985). The discovery of contemporary bacteria that can gain energy to support growth by coupling the oxidation of organic acids, alcohols, H_2, or aromatic compounds to the reduction of Fe(III) (Lovley 1993; Nealson and Saffarini 1994) support this suggestion. However, previous studies of freshwater and marine dissimilatory Fe(III)-reducing bacteria have demonstrated that, without exception, these organisms cluster in closely related phylogenetic groups in the delta and gamma subclasses of the Proteobacteria (Lovley and Phillips 1986; Rossello-Mora et al. 1994). This suggested a lack of evolutionary diversity, despite the isolation of these bacteria from such varied environments as freshwater and marine sediments, submerged soils, deep pristine aquifers, and petroleum-contaminated aquifers. If Fe(III) reduction was an early form of microbial respiration, one would expect that the capacity for coupling organic acid oxidation to Fe(III) reduction would be found in different lines of descent in the Bacteria. Thus, the available phylogenetic data are inconsistent with the hypothesis that Fe(III)-reducing bacteria played a globally significant role in the Precambrian biosphere.

In this study, we report the isolation and characteristics of a phylogenetically distinct Fe(III)-, Co(III)-, and S°-reducing bacterium which, together with *Flexistipes sinusarabici*, forms a separate line of descent within the Bacteria.

Materials and methods

Source of organism

Inocula for enrichments were collected by taking cores of a hydrocarbon-polluted ditch near Norman, Oklahoma, USA. The top 3 cm of sediment were extruded into glass tubes, stoppered, and immediately transported back to the laboratory, where they were placed in an anaerobic chamber.

Media and cultivation

Standard anaerobic techniques were used throughout the study (Bryant 1972; Balch and Wolfe 1976). All anoxic media were boiled and cooled under a constant stream of N_2:CO_2 (80:20, v/v), dispensed into aluminum-sealed culture tubes under the same gas phase, capped with butyl rubber stoppers, and sterilized by autoclaving (121° C, 20 min.). Additions to sterile media, inoculation, and sampling were done using syringes and needles (Balch and Wolfe 1976). All incubations were at 35° C in the dark.

The basal medium contained (in g l^{-1} of deionized H_2O) $NaHCO_3$ (2.5), NH_4Cl (1.5), KH_2PO_4 (0.6), KCl (0.1), and vitamins (10 ml) and trace minerals (10 ml) (Balch et al. 1979; Lovley and Phillips 1988). For enrichment of iron-reducing bacteria, a palmitic acid emulsion (13% palmitic acid and 2.7% Na_2CO_3, wt/v) was added as the electron donor (ca. 5 mM palmitic acid final conc.) and amorphous ferric oxyhydroxide (ca. 100 mM final conc.) was added as the electron acceptor. Amorphous ferric oxyhydroxide was prepared as described previously (Lovley and Phillips 1986). The enrichment was initiated by adding 1.0 g (wet weight) of anoxic sediment to sterile tubes containing the palmitate-Fe(III) basal medium (10 ml) inside the anaerobic chamber. The headspace of enrichment tubes was evacuated and replaced with N_2:CO_2 (80:20, v/v) immediately after inoculation.

The basal medium was modified to test for the use of different electron donors or acceptors. The use of different electron acceptors was tested with sodium acetate (10–30 mM) as the sole electron donor. Duplicate determinations were done for each acceptor. The concentration of Fe(II), Co(III)-EDTA or sulfide in replicates did not differ from the mean by more than 10%. Negative controls for the use of alternate electron acceptors did not contain acetate. Inocula (10%, v/v) for electron acceptor experiments were from cultures grown with basal medium, acetate, and ferric pyrophosphate [Fe(III)-PP_i] (Pfaltz and Bauer, Waterbury, Conn., USA). The use of poorly crystalline Fe(III) oxide (ca. 100 mM) and Fe(III) citrate (20 mM) as alternative reducible forms of Fe(III) was determined by monitoring the increase in cell number and Fe(II) over time. Synthetic MnO_2 (30 mM) (Lovley and Phillips 1988) was used to evaluate the potential for Mn(IV) reduction; the production of Mn(II) was followed over time. Sodium nitrate, sodium thiosulfate, sodium sulfite, sodium sulfate, sodium fumarate, or sodium malate was added to the basal medium from anoxic stock solutions to provide 20 mM. Growth on these acceptors was monitored by following the increase in cell density at an $OD_{540\ nm}$. U(VI) was provided as uranyl chloride (0.5 mM); the loss of visible color in the medium and the accumulation of a black precipitate of reduced uranium (uraninite) were monitored over time as evidence of U(VI) reduction. Elemental sublimed sulfur flower (1.0 g l^{-1}) was added aseptically to sterile medium under a stream of sterile N_2:CO_2 (80:20, v/v). Sulfide production and cell numbers were monitored over time. Co(III)-EDTA medium was prepared as described previously (Dwyer et al. 1985; Caccavo et al. 1994). The decrease of Co(III) and increase in cell numbers over time were monitored as an indication of reduction and growth, respectively. The use of O_2 as an electron acceptor was determined by measuring the $OD_{600\ nm}$ of a culture grown aerobically in basal medium with acetate and shaken at 150 rpm. The facultatively anaerobic Fe(III)-reducing bacterium *Shewanella alga* strain BrY (Caccavo et al. 1992; Rossello-Mora et al. 1994) served as a positive control.

The use of various electron donors was tested with the basal medium without acetate and with Fe(III)-PP_i as the electron acceptor. The electron donor was added to the basal medium from a sterile anoxic stock solution to give a final concentration of 10–30 mM. A H_2:CO_2 mixture (80:20, v/v; 10 ml) was added to the gas phase to test for the use of H_2 as an electron donor. Inocula (10%, v/v) for electron donor experiments were from Fe(III)-PP_i cultures that were grown in basal medium with 2 mM acetate. Duplicate determinations were done for each donor. The use of a compound as an electron donor was determined by comparing the production of Fe(II) to negative controls that lacked the compound and to positive controls that contained 10 mM sodium acetate.

The stoichiometry of acetate oxidation was determined with cultures of strain PAL-1 grown in basal medium with 10 mM Fe(III)-NTA (nitriloacetate) and 5 mM sodium acetate. The stoichiometry of proline oxidation was determined with cultures of strain PAL-1 grown in basal medium with 10 mM Fe(III)-PP_i and 0.2 mM proline.

The optimum temperature for Fe(III) reduction by PAL-1 was determined by growing PAL-1 in basal medium with acetate and Fe(III)-PP_i at different temperatures and following the production of Fe(II) over time. The ability of strain PAL-1 to reduce Fe(III) under saline conditions was determined by growing it in basal medium with acetate, Fe(III)-PP_i, and no NaCl, 0.5%, 1.0% or 2.0% NaCl, and determining the concentration of Fe(II) produced between 1 and 9 days.

DNA isolation and sequencing of the 16S rRNA gene

DNA of strain PAL-1 was isolated following the small-scale DNA isolation method of Wisotzkey et al. (1990). In vitro amplification of the 16S rRNA genes and direct sequencing of the amplified DNA fragments were done as described previously (Springer et al. 1993). The new sequence was added to an alignment of about 2800 homologous bacterial 16S rRNA primary structures using the aligning tool of the ARB program package. Similarity and distance matrices were calculated with the program SEDIS of the same package. Phylogenetic trees based on all available 16S rRNA sequences of bacteria (Maidak et al. 1994; Van de Peer et al. 1994) were reconstructed applying the distance matrix and maximum parsimony methods as implemented in the corresponding programs of the PHYLIP (NEIGHBOR) and ARB (ARB-PARS) packages, respectively (Felsenstein 1982).

Cytochrome content

The cytochrome content was analyzed from cells grown in basal medium with acetate (10 mM) and Fe(III)-PP_i. A dithionite-reduced *minus* air-oxidized difference spectrum of whole cells was obtained on Shimadzu 2101PC spectrophotometer. Cells (0.933 mg protein ml^{-1}) were resuspended in 20 mM Pipes (pH 7.0). The protein concentration was determined by the Micro Protein Assay (Pierce, Rockford, Ill., USA) with bovine serum albumin as a standard. Samples were diluted in 1.0 N NaOH and heated at 90° C for 10 min prior to the protein assay.

G+C content

The G+C contents (mean of four determinations) of the bacterial strains were determined by HPLC analysis as described by Mesbah et al. (1989) using lambda DNA as a standard. Two clostridial strains, *Clostridium cadaveri* and *Clostridium fallax*, with known G+C contents, were used as reference strains.

Analytical techniques

Fe(III) reduction was monitored by measuring the accumulation of Fe(II) over time. The amount of Fe(II) solubilized after 15 min in 0.5 N HCl was determined with ferrozine as described previously

(Lovley and Phillips 1988). Co(III)-EDTA reduction was determined by measuring the loss of Co(III) over time spectrophotometrically at 535 nm. Sulfide was measured colorimetrically (Tanner 1989). Mn(II) was measured colorimetrically by the formaldoxime method (Armstrong et al. 1979). Acetate concentrations were determined by HPLC as described previously (Lovley and Phillips 1989). Cell numbers in cultures growing with Fe(III) or Co(III) were determined by a modification of the epifluorescent microscopy technique (Hobbie et al. 1977) as described previously (Lovley and Phillips 1988). Cell density in cultures without metals were monitored spectrophotometrically at 540 nm.

Cells for electron microscopy were grown in Fe(III)-PP_i medium with acetate as an electron donor. Subsamples were removed at mid-exponential phase, stained with 2% ammonium molybdate and examined with a Philips TEM-400 electron microscope at 120 kV.

Results

Isolation of strain PAL-1

The primary enrichment was transferred six times (10% inoculum) into the basal medium with sodium palmitate and amorphous ferric oxyhydroxide. The Fe(III) in the initial enrichment tube was completely reduced within 12 days. Subsequent transfers became completely reduced within 14 days. The sixth transfer of the enrichment was streaked for isolation on anoxic Fe(III)-PP_i agar slants, which were made by adding a final concentration of 1.5% (wt/vol) purified agar to the basal medium containing acetate and Fe(III)-PP_i. Pinpoint colonies developed within 17 days. The colonies and surrounding medium turned white as the Fe(III) in the medium was reduced. Colonies were restreaked onto agar slants until a pure culture, designated strain PAL-1, was obtained. Cells from isolated colonies were vibrioid, 2–3 μm in length by 0.5 μm in width. They were motile by a single polar flagellum (Fig. 1) and stained gram-negative. Spores were not observed in wet mounts by phase-contrast microscopy. Strain PAL-1 was maintained on basal medium with acetate and Fe(III)-PP_i.

Electron donors and acceptors

Strain PAL-1 grew by the oxidation of acetate coupled to the reduction of Fe(III)-PP_i (Fig. 2). The increase in cell numbers coincided with the production of Fe(II) in the presence of acetate and Fe(III). No Fe(III) reduction or cell growth occurred in the absence of acetate. Strain PAL-1 also coupled acetate oxidation to the reduction of ferric citrate and amorphous ferric oxyhydroxide (data not shown). The theoretical stoichiometry for the complete oxidation of acetate coupled to Fe(III) reduction is as follows:

$$CH_3COO^- + 8[Fe^{3+}] + 3H_2O \rightarrow CO_2 + HCO^{3-} + 8H^+ + 8[Fe^{2+}]$$

Strain PAL-1 produced 7.86 ± 1.72 (mean ± SD, n = 3) mol Fe(II) per mol acetate oxidized, which is very close to the amount expected if acetate is completely oxidized to CO_2.

Strain PAL-1 also grew by coupling the oxidation of proline to the reduction of Fe(III)-PP_i (Fig. 3), although the rate of growth was lower than with acetate as the electron donor. The theoretical stoichiometry for the complete oxidation of proline coupled to Fe(III) reduction is as follows:

$$C_5H_9O_2N + 8H_2O + 22[Fe^{3+}] \rightarrow 5CO_2 + NH^{4+} + 21H^+ + 22[Fe^{2+}]$$

Although the proline concentration was not measured, there was a net production of 3.93 ± 0.50 mM Fe(II) (mean ± SD, n = 4) when a limiting concentration of 0.2 mM proline was added to the culture. This suggested that strain PAL-1 produced 19.6 ± 2.9 mol Fe(II) per mol of proline oxidized. This is very close to the amount of Fe(II) expected if proline is completely oxidized to CO_2.

Strain PAL-1 also reduced Fe(III)-PP_i with H_2 (101 kPa), lactate (10 mM), propionate (5 mM), casamino acids (1 g l^{-1}), succinate (10 mM), fumarate (10 mM) pyruvate (10 mM), or yeast extract (1 g l^{-1}) as the electron donor. It did not reduce Fe(III) with butyrate (5 mM),

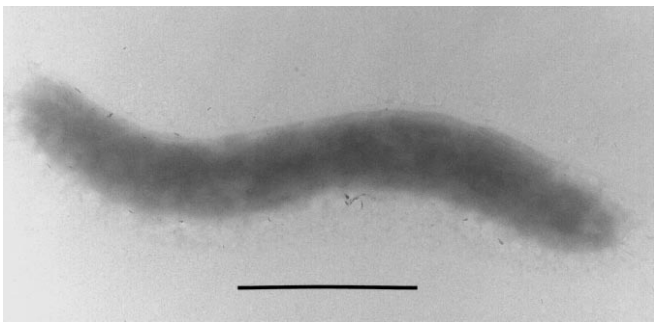

Fig. 1 Transmission electron micrograph of negatively stained *Geovibrio ferrireducens*, strain PAL-1. *Bar* 0.5 μm

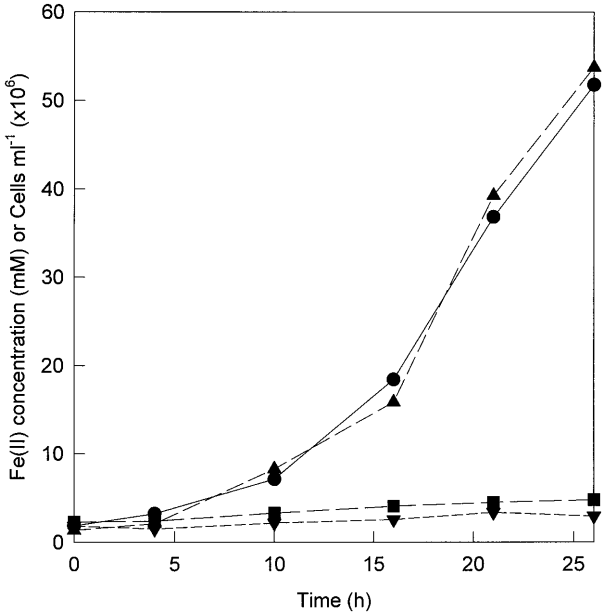

Fig. 2 Growth of PAL-1 in basal medium with acetate as electron donor and Fe(III)-PP_i as electron acceptor. Fe(II) with acetate (●), Fe(II) without acetate (■), cells with acetate (▲), cells without acetate (▼)

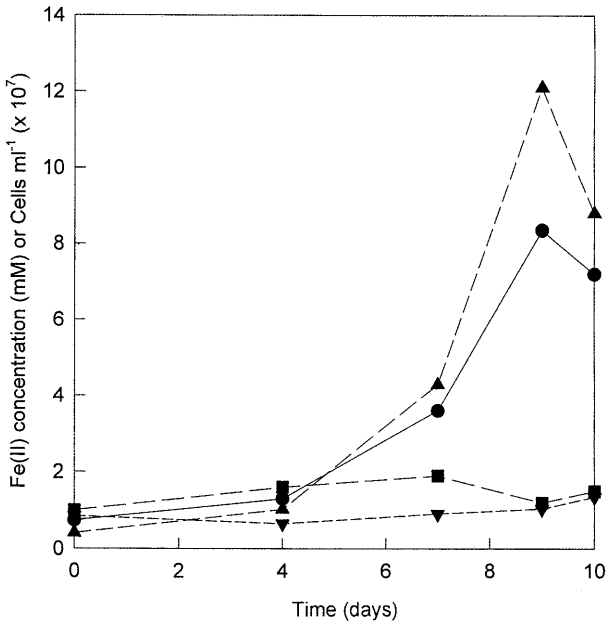

Fig. 3 Growth of PAL-1 in basal medium with proline as electron donor and Fe(III)-PP_i as electron acceptor. Fe(II) with proline (●), Fe(II) without proline (■), cells with proline (▲), cells without proline (▼)

ethanol (10 mM), methanol (10 mM), phenol (0.5 mM), toluene (1 mM), benzene (1 mM), cyclohexane (1 mM), palmitate (1 mM), benzoate (1 mM), octane (1 mM), eicosane (1 mM), phenanthrene (0.6 mM), glucose (10 mM), citrate (10 mM), nitriloacetate (4 mM), laurate (1 mM), formate (10 mM), leucine (1 mM), asparagine(1 mM), histidine (1 mM), glutamate (1 mM), or alanine (1 mM) as an electron donor. For the latter electron donors, the concentration of Fe(II) produced after 9 days in cultures with these electron donors was similar to that in basal medium without an electron donor present.

Strain PAL-1 coupled the oxidation of acetate to the reduction of Co(III)-EDTA (data not shown). Some growth of strain PAL-1 was observed in medium with acetate as an electron donor and S° as sole electron acceptor. The final cell concentrations were very low, only about 1×10^6 cells ml^{-1}. This was about 50 times less than when Fe(III) was provided as the electron acceptor. However, strain PAL-1 could be repeatedly transferred into basal medium with acetate and S° as the sole electron donor and acceptor with continued growth and S° reduction. No increase in cell numbers and sulfide concentration was detected in controls without S° or acetate. Strain PAL-1 did not reduce Mn(IV), U(VI), or Cr(IV) with acetate as electron donor. No increase in optical density was observed with oxygen, nitrate, sulfate, sulfite, thiosulfate, malate, or fumarate as the electron acceptor and acetate or lactate as electron donor.

Strain PAL-1 did not grow aerobically in the basal medium with acetate, while the facultative anaerobe *Shewanella alga* strain BrY did grow in this medium with lactate as the electron donor. Strain PAL-1 did not grow aerobically or anaerobically in nutrient broth or tryptic soy broth.

The optimum temperature for Fe(III) reduction was 35° C. No Fe(II) production was observed at 50° C. Strain PAL-1 reduced Fe(III) after 1 day in medium without NaCl. Fe(III) reduction was observed after 2 days in 0.5% NaCl medium, 4 days in 1% NaCl medium, and 9 days in 2% NaCl medium.

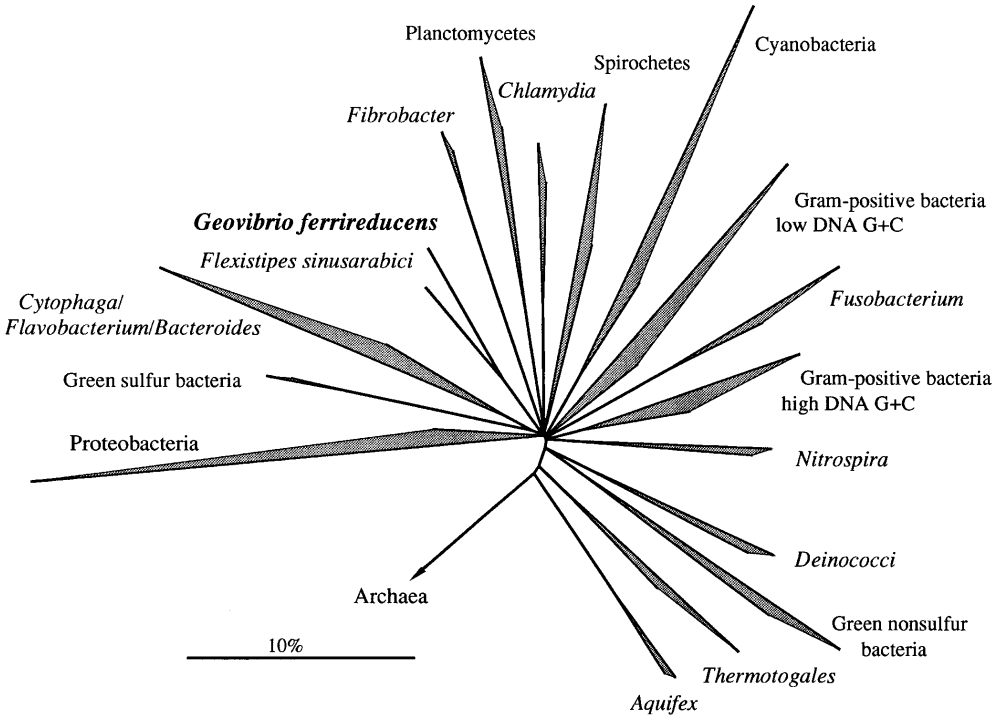

Fig. 4 Phylogenetic tree showing the relationship between strain PAL-1 and representatives of the major bacterial lines of descent. The tree is based on distance matrix and parsimony analyses of about 2800 16S rRNA sequences from bacteria. Homologous archaeal sequences were included to root the tree. The *triangles* indicate the major bacterial phylogenetic groups. Multifurcations indicate that a relative branching order could not be unambiguously determined. *Bar* indicates 10% estimated sequence divergence

Cytochromes

The difference spectra of whole cells showed the presence of *c*-type cytochromes. The reduced cytochromes had absorbance peaks at 553 nm, 523 nm, and 422 nm.

G+C Content and phylogeny

The G+C content of strain PAL-1 was 42.84 ± 1.51 mol%. The 16S rRNA sequence of strain PAL-1 was determined by direct sequencing of in vitro amplified DNA. Sequence data have been deposited at the EMBL sequence data library under the accession number X95744. PAL-1 was not related to any of the previously described Fe(III)-reducing bacteria (Fig. 4). The 16S rRNA sequence of strain PAL-1 showed 83.1% identical residues with that of *F. sinusarabici*. These two bacteria (genera) form a separate line of descent within the Bacteria.

Discussion

Strain PAL-1 is phylogenetically distinct from all other described Fe(III)-reducing bacteria. Comparative 16S rRNA sequence analysis revealed a moderate relationship between PAL-1 and *Flexistipes sinusarabici*, a flexible, obligately anaerobic, halophilic, rod-shaped, heterotrophic bacterium isolated from the Red Sea (Fiala et al. 1990). According to 16S rRNA (Fiala et al. 1990) and elongation factor Tu (Ludwig et al., 1991) sequence data, *F. sinusarabici* forms a separate phylum among the known major bacterial lines of descent. Strain PAL-1 is the second member of this phylum (Fig. 4). However, unlike *F. sinusarabici*, strain PAL-1 is a freshwater mesophile that cannot grow fermentatively. The moderate, but distinct phylogenetic relationship between strain PAL-1 and *F. sinusarabici*, and the marked phenotypic differences between these two organisms, indicate that strain PAL-1 represents a new genus within this phylum. We propose that strain PAL-1 be established as the type strain of a new genus and species, *Geovibrio ferrireducens*.

Although PAL-1 was enriched from sediments with palmitate as an electron donor, it did not couple palmitate oxidation to Fe(III) reduction. It is likely that another organism in the primary enrichment converted palmitate to acetate, providing a substrate for strain PAL-1.

The complete mineralization of complex organic matter involves the concerted action of several metabolic groups of bacteria (McInerney and Bryant 1981; McInerney 1986). In methanogenic ecosystems, fermentative bacteria, syntrophic bacteria, and methanogenic bacteria are required. In marine environments, the functions of the syntrophic bacteria are performed by sulfate-reducing bacteria (McInerney 1986; Widdel and Hansen 1992). In Fe(III)-reducing environments, a complex trophic structure also exists (Lovley 1991). Fermentative bacteria hydrolyze complex substrates such as proteins, lipids, and polysaccharides and ferment sugars and amino acids to acetate and longer chain fatty acids, H_2, formate, CO_2, NH^{4+} and HS^-. The fermentative bacteria may also reduce a small amount of Fe(III). The members of the genera *Geobacter* (Lovley and Phillips 1986; Lovley et al 1993a), *Desulfuromonas* (Lovley et al. 1993b), and *Desulfovibrio* (Lovley et al. 1993b) couple the complete oxidation of acetate to Fe(III) reduction. These organisms are all members of the delta subclass of the Proteobacteria. Until recently, only members of the gamma subclass of the Proteobacteria, *Shewanella* and *Pseudomonas* species (Balashova and Zavarzin 1980; Lovley et al. 1989; Caccavo et al. 1992; Rossello-Mora et al. 1994), were known to use H_2 and formate as electron donors for Fe(III) reduction. These organisms can also incompletely oxidize lactate and pyruvate. However, *Geobacter sulfurreducens* (Caccavo et al. 1994), a member of the gamma subclass of the Proteobacteria, and now *Geovibrio ferrireducens*, have been shown to use either H_2 or acetate as electron donors for Fe(III) reduction. The complete oxidation of several aromatic compounds by *Geobacter metallireducens* and the ability to use propionate, a key intermediate in anaerobic decomposition, by both *G. metallireducens* and *Geovibrio ferrireducens*, indicates that the degradation of fatty and aromatic acids in Fe(III)-reducing environments is possible without syntrophic associations, as found in methanogenic environments.

Strain PAL-1 is the first bacterium described in pure culture which can utilize an amino acid, proline, as a sole source of carbon and energy for Fe(III) reduction. While *Shewanella putrefaciens* strain MR-1 requires arginine, glutamine, and serine for cell synthesis, lactate, formate, pyruvate, or H_2 are used as the electron donor for Fe(III) reduction (Lovley et al. 1989).

Like *G. sulfurreducens* (Caccavo et al. 1994), *Desulfuromonas* species (Pfennig and Biebl 1976), *Desulfuromusa* species (Finster and Bak 1993), and *Pelobacter* species (Lovley et al. 1995), strain PAL-1 coupled the oxidation of acetate to the reduction of S°. This provides further pure culture evidence supporting the interrelationship between the microbial iron and sulfur cycles in anoxic environments (Coleman et al. 1993; Perry et al. 1993; Roden and Lovley 1993; Caccavo et al. 1994).

Geovibrio ferrireducens strain PAL-1 is an important addition to the rapidly growing group of respiratory metal-reducing bacteria available in pure culture. More than 30 isolates have been shown to gain energy for growth by coupling organic matter oxidation to dissimilatory Fe(III) reduction. These organisms now include members of the genera *Geobacter* (J. D. Coates et al. unpublished results), *Desulfuromonas* (J. D. Coates et al. unpublished results), *Desulfuromusa* (Finster and Bak 1993), *Pelobacter* (Lovley et al. 1995), and *Wolinella* (D. R. Lovley et al. unpublished results). However, all of these isolates are members of the Proteobacteria. *G. ferrireducens* expands our concept of the types of bacteria which may contribute to organic matter mineralization and Fe(III) reduction in anoxic environments, and illustrates that the capacity for dissimilatory metal reduction is not only restricted to members of the Proteobacteria. The

ability to use Fe(III) as a terminal electron acceptor is now found in more than one line of descent in the Bacteria, which is consistent with the concept that Fe(III) reduction played an important role in the Precambrian biosphere.

Description

Geovibrio (gen. nov.), Ge.o.vi'bri.o Gr. n. geo the Earth; L. hyp.masc.v. vibrare to vibrate; N.L. masc.n. *Geovibrio* vibrating from the earth.

Vibrio-shaped, gram-negative cells, 2–3 × 0.5 μm, motile by single polar flagellum, without spores. Cells occur singly or in chains. Strictly anaerobic chemoorganotroph that oxidizes acetate with Fe(III), S^o, or Co(III) as an electron acceptor. Hydrogen, proline, lactate, propionate, casamino acids, succinate, fumarate, pyruvate, and yeast extract are also used as an electron donor for Fe(III) reduction, while other carboxylic acids, sugars, alcohols, amino acids, and aromatic hydrocarbons are not. Growth in the absence of an electron acceptor was not observed. Temperature optimum 35° C. Cells contain *c*-type cytochromes. Grows best in freshwater medium, but will grow in up to 2% NaCl.

Geovibrio ferrireducens (sp. nov.), fer.ri.re.du'cens L.n. ferrum iron; L. part. adj. reducens converting to a different state; N.L. adj. *ferrireducens* reducing iron.

The description is the same as for the genus.

The type strain of *Geovibrio ferrireducens*, PAL-1 has been deposited in the American Type Culture Collection under ATCC 51996. *Geovibrio ferrireducens* was enriched from surface sediments of a ditch in Norman, Oklahoma.

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